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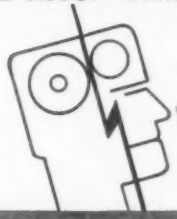

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FT food technology

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Facts About Food Supplies

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In giving permission to FOOD TECHNOLOGY to reproduce this article, Professor Nicol wrote, "Food is a chemical substance or set of substances. Its production and use require to be considered in accordance with the reality of established principles of chemistry and nutrition. Appreciation of the circumstances governing production of food can come only by going along with the facts—not by being at odds with them." In urging an appreciation of the potentials of basic chemistry for interpreting and leading the way to the solution of food problems, Professor Nicol registers a strong protest against present-day public apathy toward the study of food science. In democracies the lay public has much to do with where scientific emphasis shall be placed and how much financial support a given area, such as food, shall receive. This democratic prerogative is as it should be except, as Dr. Nicol points out, "only those gaps (in scientific curricula) about which laymen are conscious are likely to be filled without much effort in argument and persuasion by scientists." In a day when space science has captured the lay public's imagination, the need for extending food science study is in some danger of being eclipsed.

At the time of the Quemoy blockade the *Daily Mail* included a special section for gastronomes and justified it by saying that "Nothing seems to stop the craze for Chinese food." About this *Punch* remarked: "Especially if you're Chinese."

The apt remark to all this seems to be something like: "Aren't we all?" Chinese or not, inside or outside the Food Group, we are all interested in food—and particularly in getting enough of it. Yet much of what is widely believed about the origins of food bears little relation to the truth. However, when the scientific facts about food are assembled, they can be seen to follow a few general but incontestable principles. There are hardly more than half a dozen basic principles; being truly basic, these principles apply to both land and sea without modifications other than local ones due to circumstance.

Dr. Hugh Nicol

Author of the accompanying article, Hugh Nicol, Ph.D., F.R.I.C., F.R.S.E., is Professor of Chemistry and Head of the Department, West of Scotland Agricultural College, 6–7 Blythswood Square, Glasgow. This distinguished essay was published originally in Chemistry & Industry, *for August 22, 1959, and appears in* Food Technology *by kind permission of that journal and of the author, Dr. Nicol. The paper was presented before the Nutrition Panel of the Food Group of the Society of Chemical Industry, April 1, 1959.*

All food comes from plants. Sometimes food plants are without chlorophyll, but photosynthesising green plants are the source of all food for all animals, including fish, and also of organic nutriment for fungi and other saphophytes.

The bulk nutrients contributed by plants are carbohydrates, fats and proteins. For brevity I will do no more than mention vitamins and mineral constituents and will consider fats along with carbohydrates to avoid the awkward phrase "non-nitrogenous constituents." So there is the simple contrast of carbohydrates (in that sense) with protein.

The next step, therefore, is to consider the nutrition of plants. This is cyclic; and the cycle—the life-cycle, that is—can be conceptually entered at any point. Wherever the cycle is begun, it must be borne in mind that we are dealing with a cycle; the round must be completed so as to take all factors into judicious consideration.

It is usual to begin with solar energy and photosynthesis. There is nothing wrong with making a beginning there; yet one of the most deeply-rooted misunderstandings about food takes rise in the imperfect teaching that begins with photosynthesis, over-emphasises its importance by calling it the focal circumstance of life (or some such nonsense) and fails to give due weight to the other equally important links in the plant-soil life-cycle. The student remembers little more than the insistence on photosynthesis and carbohydrate.

One could embroider the pattern of Figure 1 extensively, say, with specialities of the nitrogen cycle; but we are here concerned only with first principles to which such things are accessory. Two primary adjuncts of the life-cycle are:

(1) All fertilisers and manures without exception either are salts directly soluble in water or become ionic through biological and other processes occurring in the soil. Chalk becomes calcium bicarbonate; farmyard manure and other "organics" are broken down by microbial action into simple ions. Unless converted into simple ions such as those mentioned in the life-cycle diagram (Figure 1), no fertiliser could be of use to plants.

(2) The life-cycle results in an accumulation of soil organic matter loosely called "humus". This has or attains some equilibrium-value in its ratio of total carbon to total nitrogen, so that the C/N ratio of soil is a factor determining productivity. Manipulating the C/N ratio of soil is a prime feature of soil management and cropping.

The prime things about food for any animal are that the day's diet must contain a certain minimum of protein, and that that sufficiency of protein must be maintained, roughly three times a day, for the next day, week, year and decade as long as the animal

lives: and for as long as we human beings expect to exist as a community.

That necessary minimum proportion of protein is, roughly, 16% or, say, a sixth of our food. To discuss the proposition properly we would have to take digestibility and biological quality into account; but I must take those for granted.

Cereals, which planners and publicists of the potentialities of fertilisers usually invoke almost exclusively (perhaps because the tale of Joseph and his brethren is void of specific reference to protein), have insufficient protein. By themselves cereals cannot sustain life; the ancient Egyptians, for instance, had fish and lentils, and possibly their barley was unusually rich in protein. In general, a human diet that does not contain either legumes eaten as such (as pulse or otherwise), or protein-rich animal products from land or sea or river, leaves an unbridgeable gap between its content of protein and the vital minimum.

There are only two ways of gaining protein from plants to supplement the low proportion obtainable from cereal grains and other mature parts of non-leguminous plants. These are:

(1) Eating any *young* plant (including yeast and the like) or newly-formed part of any green plant;

(2) Eating any part of a nitrogen-fixing leguminous plant (legume) at any stage of growth.

In those two ways we keep alive. Unlimited photosynthesis alone—artificial or other—will produce only

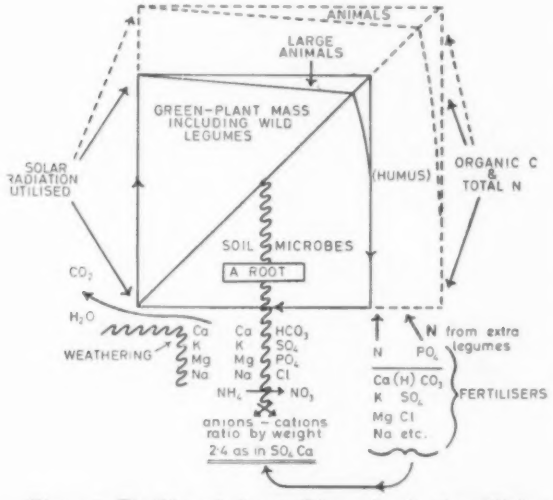
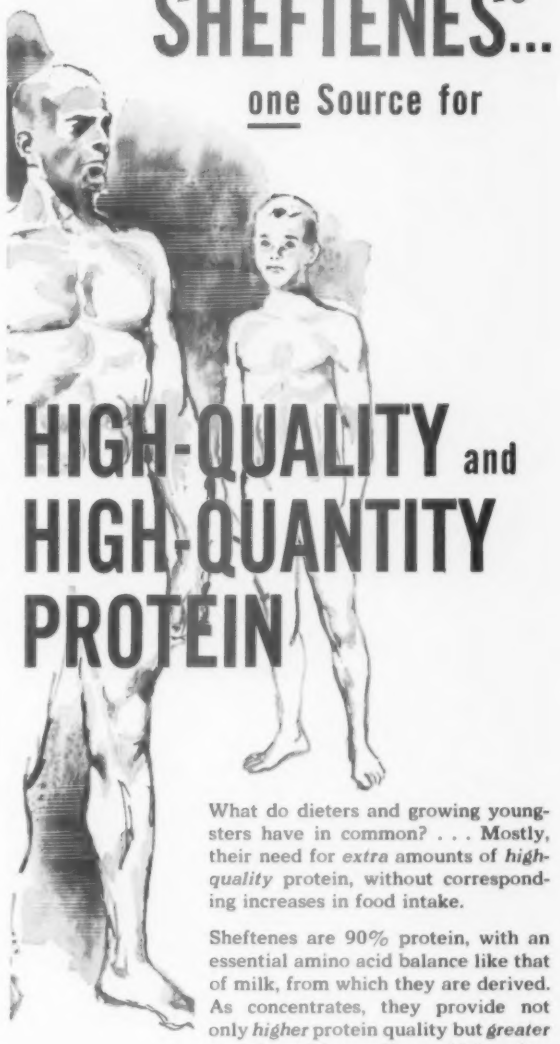


Figure 1. The life-cycle for a soil in good natural equilibrium (full lines). Photosynthesis induces incorporation of nitrogen from soil into plants, and supports animals accordingly. Inorganic compounds of nitrogen cannot easily produce food capable of sustaining animals; but nitrogen fixed biologically in leguminous plants supplied with non-nitrogenous fertilisers—especially phosphates—enlarges the cycle (dotted lines), permitting proportionately more animals (on two legs and four) to remain alive. Microbial decomposition of once-living matter, supplemented by rock weathering, produces CO_2, H_2O, and ions—taken up by plants. In a normal productive soil the overall ratio of anions to cations will be 2·4: 1 by weight. Intelligent application of fertilisers and other materials to soil: (i) augments the natural stock of "nutrient" ions; while (ii) bringing or maintaining the anion-cation weight-ratio near to 2·4.



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sugar or carbohydrates; it will never help unless we can *also* find means for harvesting the protein-rich growths of young non-legumes or a sufficient growth of legumes.

For increasing the food supply, greater use of fertilisers is the stock recommendation—or should I say, the talisman? Nitrogenous fertilisers loom large (especially with chemical apologists).

Consider nitrogenous fertilisers first, then, for the facts about their relation to food. All nitrogenous fertilisers and manures, without exception, have features in common besides ionic effects shown by all fertilisers. They also have special features depending on the way in which their nitrogen is combined. If any nitrogenous fertiliser—or, for brevity, just "nitrogen" to be understood as "nitrogen in combination as any fertiliser"—is used, it does three things:

(1) It temporarily narrows the C/N ratio of soil; and

(2) It promotes photosynthesis. It does that, not by raising the efficiency of photosynthesis per unit of area, but by increasing the photosynthesising area through making green plants grow larger. Giving extra nitrogen, then, is an effective way of increasing photosynthetic output or capture of solar radiation by plants; it is, indeed, the only method so far discovered for doing that. This effect is really another aspect of C/N balance, but within the plant.

There are some pendants to this enlargement of photosynthesis.

(a) If the plant is harvested when it is mature (as cereals and potatoes or turnips may be said to be), the increased photosynthesis closely matches the fertiliser-N supplied, with the result that there is a greater total weight of protein per acre and a correspondingly larger yield of carbohydrate (i.e. C/N ratio constant, roughly). There is thus more "food," of sorts unsatisfying by themselves; and nothing has been done to bridge the protein gap. The gulf to be bridged has, instead, been widened—because still more protein is now required to give nutritional balance to all that extra carbohydrate. As Lawes and Gilbert stated, a characteristic of nitrogenous fertilisers is to increase the output of non-nitrogenous constituents—in non-leguminous crops harvested mature, is what they meant.

(b) If a plant is caught very young—with not more than a few weeks' growth—it will have converted the nitrogen at its disposition (whether from fertiliser or native to its habitat) into protein in amounts at least as rich as any legume at any stage of growth. That is the origin of fish protein and of proposals for culturing yeasts or microscopic green algae for food; it is the basis of rotational grazing of grassland, of grass-drying and some other intensive forms of grassland agriculture (being supplemented by pasture legumes); it also accounts for the relative richness in protein—albeit of second quality—in cabbage, brussels sprouts and the like.

This kind of feeding occurs naturally in the sea, where there are no legumes. It can be made use of in practical farming only through appeal to pasture or

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grazing kale; but wild quadrupeds often benefit from it by grazing young shoots of trees.

Securing young growth of plants implies frequent harvesting. This can be done mechanically—as in production of dried grass or microscopic algae—or by intensive grazing. Nitrogenous fertiliser applied to any such young growth does produce a high proportion of protein; but the problem of protein-production is not thereby solved. A limit is set by the length of the growing period, which commonly does not exceed six months. The problem remains of keeping animals alive on protein preserved for the other half of the year. At this stage it seems to be enough to state the problem—which is a real one. Practically the only place in the world where grazing is possible the year round is New Zealand—having less than one-thousandth of the world population.

(c) It is normally uneconomic to give nitrogenous fertiliser to legumes in bulk (i.e., in virtually pure stand, unmixed with true grasses).

(3) Nitrogenous fertilisers imperatively set up a requirement for *metallic* base. It is essential to correct the ionic disturbance, including acidity, invariably elicited in soil when a nitrogenous compound free from a base-forming metal is used. For neutralizing acidity it does not matter how the base is supplied—whether chemically combined as in sodium or calcium nitrate or mixed-in as chalk added or naturally present in the soil—but the needs for base other than ammonium are inescapable when any form of combined nitrogen is used. It follows that a programme for artificial fixation of nitrogen into fertilisers must be matched by provision of a much larger tonnage of metallic alkalies or carbonates.

To return to paragraph (1) about "nitrogen"; the C/N ratio of soil might be called the "law of gravity" of soil biochemistry. Like the need for metal base along with nitrogen, there is no getting away from it. Textbooks usually mention the C/N ratio in a detached way in odd corners, as if it did not matter very much. Food-and-population theorists never mention it.

Nitrogen is the inseparable concomitant and ballast that trims photosynthesis. However the nitrogen is combined (whether as a result of artificial or biological fixation) it governs photosynthesis in two modes. One is in the plant itself: increased nitrogen permits more leaf area and plant bulk, as already pointed out. The other controls decomposition in soil and sea; it completes the life-cycle and decides its rate of revolution.

If—as some dreamers hope—photosynthetic output were doubled through some incantation, in the name of "science", to solar energy to be captured by green plants without more combined nitrogen being found, the C/N ratio in plants and soil would be doubled; bacterial breakdown in the soil would be slowed so that the bigger, but nutritionally useless, plants could be grown about half as often as now; the sea and rivers would probably stink. Not that that would matter to us, since we should die from being deprived of our present narrow margin of balancing protein. Adding extra fertiliser-nitrogen to soil would correct its C/N ratio and restore the rhythm of plant life; but it still would not feed our present numbers. Bacterial fixation of atmospheric nitrogen in legumes does not just provide bridging protein; it also takes care of the C/N ratio in the soil. Surely the facts are much more interesting than fancies could ever be!

I sometimes tell students that not even in agriculture can they get something for nothing. The rule of Nature is "nothing for nothing, and precious little for sixpence." To get more food—real food, that is, containing a due proportion of protein—we must pay a cost in fuel: chemically speaking, in reducing substances. So, apart from questions of adequacy of land, or sudden catastrophe, the future of man will be decided by the ability to have enough fuel at all times to feed enough legumes and other crops and to collect enough fish, to be in nutritional correspondence with whatever population there may be. That is a condition of existence henceforth.

Most bridging protein comes from ordinary phosphatic, nitrogen-free fertilisers put to sustaining leguminous crops. Use of phosphate—along with potassium salts as necessary—can enable clovers and other legumes to supply enough nitrogen for most growth-promoting purposes of grasses and livestock. The prime example is New Zealand, where nitrogenous fertilisers are little used except for cereals and fruit.

Potassium fertiliser, according to vague advice in the textbooks, is needed to "balance" N as plant nutrient; the more fertiliser-N used, the greater the requirement for K, as a rule. For the moment, I am deliberately following the shocking, but hitherto world-wide, practice of allowing chemical names or symbols of "nutrient" elements to stand for whole salts. It is common to recommend or specify (if that is the word) K or potassium or potash, without regard to the anion and its effects; and *mutatis mutandis* for other fertilisers.

If potassium chloride is used in high concentration, the books do recognise that it produces something called "chloride toxicity." There is no warrant, here or elsewhere, for implicating a single ion rather than its partner in the same salt, but Cl is not recognised as a "nutrient," so that it must share with Na the odium of being "nasty" or "toxic"; and, for high-value crops such as tomatoes, the sulphate of potassium is preferred to the chloride. On the exclusive nutrient theory about NPK, which takes no account of the ways in which simple salts actually behave, the absence of chemical principle leads to obscurities and confusion.

A result of these unnatural complications is that many people—market-gardeners, for instance—understand tigers (which do not come into their experience) much better than they understand the fertilisers that form a large part of their business. It would be ridiculous to visualise a tiger as nothing more than a set of jaws and teeth; the legs, at least, must come into the picture: and, in fact, the whole animal. Yet market-gardeners and farmers and their advisers

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are often content to evaluate fertilisers in terms of some nutrient—K or P or N—as if that were the "business" end of a fertiliser and nothing else in its make-up mattered. Since the pictures of fertilisers are misleading, is it to be wondered at that crop-producers are frequently puzzled by the simplest fertilisers, as when they do what they are told and use a lot of "K" to feed their plants generously and still have troubles about the nutrition and well-being of their crops?

For example, fertilising tomatoes with large doses of K has to be balanced with more Mg than is needed as nutrient; and, since K and Mg are both supplied as sulphate, there is a tremendous dose of SO_4, which, on the nutrient theory, it is convenient to ignore. In that way "toxicity" wrongly attributed to chloride or Cl is averted; but the fruit may be badly shaped or cracked through exposing the plants to unnatural and unrecognised ionic stresses.

Mr. G. D. Lockie, of Plant Protection Ltd., has avoided much of this trouble by defying convention about K as "nutrient" and empirically replacing some of the potassium sulphate with sodium sulphate. That is ionically sound, because it restores the anion-cation weight ratio more nearly to normal, whereas a high proportion of potassium sulphate or chloride lowers that ratio to a pathological point. Mr. Lockie's use of sodium sulphate exemplifies a rational method of cheapening some fertilising, yet improving or maintaining yields and commercial quality.

One of the hoariest legends about food is that about the feeding of the urban population of Britain after the Industrial Revolution by cereals from overseas. That is nutritionally incorrect. The same sort of notion is held by F.A.O. and its followers in reckoning surplus wheat and sugar among increases of "food" production.

The immediate explanation for this aberration of F.A.O. about world food is that the Organisation is dominated by economists who concern themselves only with *commodities*, i.e. what is potentially saleable; they thus overlook the crucial forage and pasture legumes which sustain animals (= bridging protein) but do not normally enter into trade. The followers of F.A.O. do no better.

It is hard to understand why no chemist or nutritionist in F.A.O. or elsewhere has acquired enough influence to spread correct ideas about food and what makes man tick; this in spite of the fact that knowledge about protein-carbohydrate ratio in food, effects of nitrogenous fertilisers, ionic behaviour of solutions, has been established for at least 50 years.

It is by an upsurge of leguminous plants, and of animals fed on their bridging protein, that the Western peoples have sustained the greater part of their increases of population since about 1840 at least. That is a matter of history, and specially the history of white colonisation of the "new" lands like those of Australasia and North America. To it must be joined, since about 1890, the circumstance of amassing food by burning fuel. The recent "advance of the legumes" makes a fascinating story; and I can only regret that, like other bases of nutrition, it has failed to obtain historical and contemporary attention so as to substitute facts for legend about food.

There are still many peoples—the Indians and Chinese, for example—who do not use appreciable amounts of fossil fuel either as fertilisers or for cultivation. Japan is an especially interesting subject to discuss. Japan is the world's heaviest user of fertilisers per acre of cultivated land. She has little mechanical cultivation or animal husbandry and uses fertiliser—that is to say, fuel—mostly for rice. So far, we seem to be in line with those who write mainly about cereals. Some beans are grown in Japan, but most of Japanese bridging protein is supplied by fish.

Recent British information about Japanese agriculture comes entirely from the writings of an employee of a chemical firm; the knowledge given is naturally scrappy and tells us little about diet and practically nothing about fish.

F.A.O. is an invaluable source of information of a commercial kind; if you want that sort of unscientific information, F.A.O. is your indefatigable squirrel for collecting and publishing it; but it lacks a scientific aim or ambit. It will tell you, for example, that there are 800 million cows in the world, and in what countries, but not how they (and you) are fed; it will give you the total tonnage of Japanese fish or rice, without a hint as to how much fuel is consumed to acquire either, or even that fuel matters at all in food production today.

In Japan we have a simplified instance of our British dependence on fuel for food; I suggest that the United Kingdom should send a study mission to learn in detail the lesson about the Japanese application of fuel and chemistry to food production for an over-populated set of islands.

In tracts about world food problems we get tables of output of cereals, potatoes and sugar alongside tables of meat output, but without a hint to say what the connexion (if any) is. Not only is the vital leguminous link effectively obscured from the public, but the myth about the supreme importance of cereals is reinforced by obtaining the sanction of "authority".

The cereal "fixation" common to accredited publicists illustrates the poor quality of much comment about food problems. It is reason for pessimism that of the "authorities" on world food supply and prospects, none seems to be able to do the most elementary homework about protein, irrigation or fertilisers.

Let us come back to fertilisers as salts. The chemistry of soil management for cropping boils down to control of ions. Any good soil has its anions and cations in such quantitative balance that their ratio, by weight, in a suitable aqueous extract of soil is not less than 2·4:1. We owe that recent finding to my colleague, Mr. E. K. Schofield-Palmer, and I must ask you to accept it. There is no time to amplify it beyond remarking that plant physiologists interested in nutrient culture have for a century empirically evolved solutions with anion-cation ratios of that magnitude.

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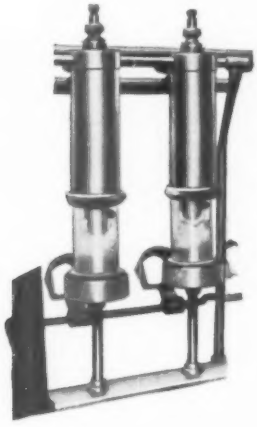

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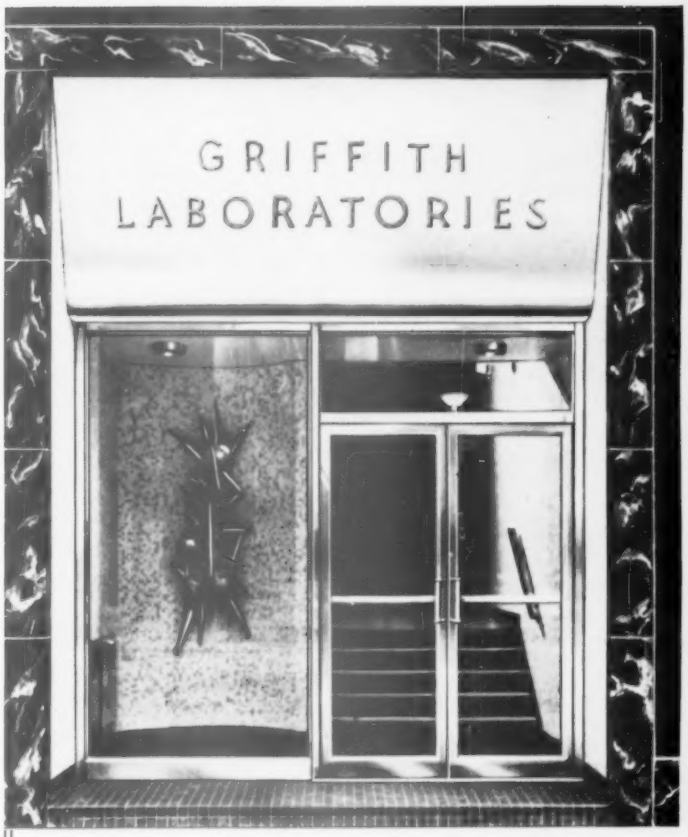

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ments, such as lime, are two sets of facts and consequences, namely:

(1) Because they ionise and set up osmotic pressure, and decrease free energy of water, they tend to oppose growth of plants (as does the native soil solution, also salts that are not regarded as fertilisers);

(2) Because they supply elements and groups called "nutrients," they tend to promote plant growth.

It will be immediately grasped that study of plant growth in relation to salts is a problem of equilibration or balance: notably, ionic balance. Most soil phenomena are equilibrium phenomena and deserve to be treated as such. Among the penalties of conventional plant-nutrient theory is its disregard of equilibria in the soil; notice has been given to equilibria only in rather unphilosophical forms like the occurrence of diminishing yield-increases from successive equal fertiliser increments. Food-production chemistry has long been approached at the technical, rather than a scientific, level.

An essentially pragmatic approach to crop research and food production is not wholly the fault of present-day fertiliser chemists. During the last hundred years—that is, since universities lost interest in the fundamentals of crop production—research in agronomy has been expected to be consistently "practical"; the emphasis on immediate aims and quick results has been a dead hand pressing so heavily on crop investigation as almost to extinguish curiosity about the ways in which plants grow, and fertilisers act, in chemical terms. Instead, there has grown up a jargon—such as about N, P, and K—which repudiates chemistry and effectively denies that fertilisers are salts.

The fertiliser cupboard has come to be a kind of ark containing some good animals, such as phosphate, potassium and nitrogen, and, it may be added, trace elements; but into the ark have somehow slipped some rather bad animals, such as chloride and sodium; the status of sulphate and of sodium is uncertain and varies on occasion from "good" to "bad" and sometimes, as we have seen, "needn't be noticed at all." Nature does not work along those lines: every ion of every salt in every solution has an effect on plant or muscle tissue or whatever it comes into contact with, and that ionic effect is neither good nor reproachable, but just natural. Each ion and its effect exists, whether nutrient and biochemical theory chooses to acknowledge it or not.

It has been usual, for instance, to ignore the calcium in most fertiliser phosphates and to ascribe their whole effect to P. "N" is allowed to stand for nitrogen irrespective of its mode of combination in particular substances, though first-year chemistry would indicate a difference between ammonium sulphate, sodium nitrate and urea. The absurdities that this leads to can be seen by reading the literature of fertilisers; chemical nonsensicality about fertilisers has even managed to blunt that otherwise logical tool—statistical analysis. Such pseudo-chemistry is unworthy of the industry of food.

A modern approach to soil and fertiliser effects in relation to plant growth is possible and comprehensive. The ionic aspect of salts in solution is surprisingly easy to master (as my colleagues have shown) by going back to first principles.

We have to deal simply with ions. These carry an electric charge, therefore their effects can be summed up in electrical conductivity or resistance. Not only "can be", but already have been—by Nature herself for every situation.

So we cannot go wrong by measuring total ionic effects (whether in hindrance or help of plant growth) by making the easy determination of electrical resistance of, say, a water extract of soil before or after any treatment such as fertilising or liming. That gives a number in ohms; but the properties of a solution containing ponderable salt ions, such as sulphate, chloride, calcium and so forth, are modified by the pH. That is measured in volts, and expresses the ratio of the two ions of the solvent, water.

Since pH is a negative logarithm, let us put specific resistance into similar form and call it pC. Those two terms can be integrated into a useful function of the physico-chemical effect of all ions from solvent and solute alike. This omnibus integral is called pL; for soil it is 2pH—pC.

pL expresses the physical effects of all ions in any solution such as soil extract or irrigation water. The chemical effects of the ions are summed in the new parameter already mentioned (Figure 1)—the ratio, by weight, of all anions to all cations. For convenience, we call that ratio, G. With pH, G gives the *distribution* of the ions; from the data for establishing G, we have all chemical information about the ions; pC gives their total *concentration*, or, more exactly, what might be called their concentration-effect as it is expressed in free energy or pF.

With that chemical and electrical information, it can be literally calculated how plant roots (for example) "feel" about any ionic environment, and, consequently, how a plant is likely to react and grow if external factors, such as temperature, are tolerable.

G tells us fully and frankly about nutrients and non-nutrients alike, while avoiding artificial distinctions about what may or may not be admitted as such according to some fashion of the time.

Excepting a human being imperfectly primed with ionic theory and its implications, no organism can pick and choose particular ionic effects, or ratios between selected pairs of ions. It is therefore clear that the biologists' concept of "antagonism" between Ca and K, or any other pair of ions chosen from a larger group, enshrines a fallacy—but it is a fallacy that has become an article of faith for many "fringe" users of chemistry.

An up-to-date definition of the purposes of soil management for producing food or other crops thus comes by joining first principles about ions to what is known or may be discovered about nutrients (however classed). It is:

To supply needed nutrients while keeping pL at an optimum not greater than about 13 and maintaining G at or slightly above 2·4.

Aluminum Foil Packaging– Advantages of High Thermal Conductivity

One of a Series of Reports by Reynolds Metals Company

Thermal conductivity is a property often overlooked or considered of little consequence in the selection of packaging materials. Growth of the frozen and refrigerated foods industry, however, is placing increased emphasis upon the thermal properties of the package. Frozen foods producers are interested in faster and more efficient freezing. Grocers are interested in obtaining maximum efficiency from storage cabinets – which are frequently overworked. And, of course, the ultimate consumer is interested in rapid thawing or heating. The high efficiency of aluminum foil as a conductor of heat makes it unique among flexible packaging materials.

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Still other benefits are to be gained in storage situations where it is important to minimize temperature fluctuations. As an illustration, the accompanying graph shows the temperatures of ice cream in pint packages of waxed fiberboard cartons with and without aluminum foil overwraps.

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These packages were stored in an open top refrigerated display cabinet typical of those generally found in self-service markets.

It is a well documented fact that ice cream deteriorates rapidly through softening, shrinking and coarsening of texture if the temperature rises even a few degrees above its proper storage temperature. The most severe test of the packaging material, naturally, takes place in the top layer of the storage cabinet since exposure to radiant heat and convection currents is greatest at that point. Practical experience shows that ice cream in this location does, in fact, deteriorate most rapidly.

Test results shown in the graph indicate that, in this most exposed layer, the ice cream in foil wrapped cartons was held at 9°F *lower* temperature than ice cream in plain cartons on the same layer.

Even in the bottom layer, where radiation and convection are minimized, the temperature of foil wrapped ice cream was maintained at 4°F colder than ice cream in plain cartons.

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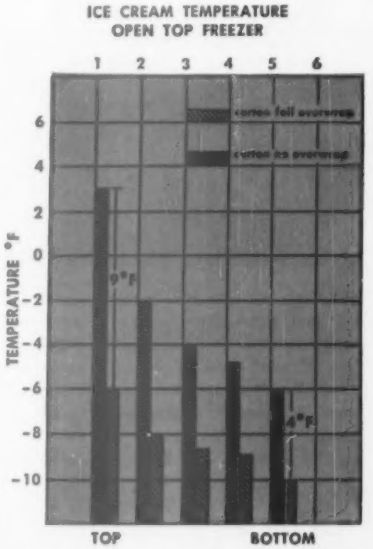

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For a solution intended for growing ordinary land plants that definition or prescription covers everything; in actual soil, the rate or intensity of release of phosphate provides a complication not yet resolved. Without giving details of fertiliser action, I am content to show that any consequence of using or not using fertilisers can be evaluated in natural ways consistent with ordinary chemistry; and that quantitative principle can come into its own throughout the sphere of production of food and crops, and probably in other spheres of chemistry too.

There is an important ionic principle with chemical interest everywhere. You have seen the undesirability of taking ratios between any number of ion-species fewer than the total present; but, if you take all ions—as you should—and calculate in equivalents (as is common in biological work), you invariably get the ratio 1·00. That produces no information; unless to show that your analysis is complete.

Accordingly, I suggest that in biological respects Nature cannot operate in terms of ion-equivalents (beyond satisfying their inevitable 1:1 ratio); but she must operate in ratios of all cations to all anions by *weight*. Therefore, disturbance of quantity of one or more of the kinds of ions in poor or over-manured soil, or in bodily disease, will have different types of effects—according as G is allowed to vary (as in soil) or is normally held constant (as it probably is in healthy muscle or blood of a given species). That idea opens up wide possibilities, provided, of course, that all ions are recorded, and not merely some of them.

Fertilisers require fuel, or at least a good deal of energy, in their making. A tremendous change supervened in man's relations with his world when fertilisers came into general use towards the end of last century—roughly about 1890, or 70 years ago.

That was a time of discovery and important innovations. All of them—except the unprecedented new note in our food-supply—have been amply publicised so that everybody has at least heard of them. *X*-rays, radiochemistry, quantum theory, the aeroplane, the steam turbine: anyone can easily add others. Those in engineering and applied physics will probably come most readily to mind; so may I add a chemical novelty not widely recognised, on which much of our protein has hung since 1904—namely, the exploitation of large sulphur deposits by the Frasch process?

Sulphur, used to make immense amounts of superphosphate, typifies the radical departure from all that had gone before in food-gathering. That has had no publicity as far as I am aware; and the "authorities" about world population and food supply do not seem to have even noticed that anything has changed about the winning of food since the time, not long ago, when all people got all their food from the annually renewable resources of sun, soil, wind and rain. No professional sociologist, economist or historian has noted the significance of the change that occurred when man began to use fossil fuel to produce and gather food. The prevalent idea still is that all food comes from current photosynthesis alone. That is a relic notion surviving from a former age.

As a loose way of expressing man's mode of living up to about 1890, it might pass muster; but it is not true, on any count, of our present circumstances or of the immediate future. To make a statement about food that shall be true, now and henceforth (as for the past 70 years or so) it is necessary to take into account both our daily income of solar energy and the energy of those reserves of fossil fuels raided to produce more food than we could get from current income alone.

The increase in population of the technically-advanced peoples for the last few decades has been upheld by crops and fish harvested by the aid of fossil fuel. Among these new and enlarged crops the legumes have been vital and central.

First about 1890 steam vessels began to replace wind-power in fishing; later came oil-driven fishing vessels, then motorised cultivation of land. There are many other mechanical and physical instances of this use of fuel. Reserves of phosphate rock and of potassium salts are ample for the foreseeable future; but the position about fuel itself is less clear. If fuels were well distributed geographically and were used only for food production, there would be no need to worry; only a small proportion of fuel is so used. However, by using fuel for non-food purposes such as going about in motor-cars and aeroplanes,

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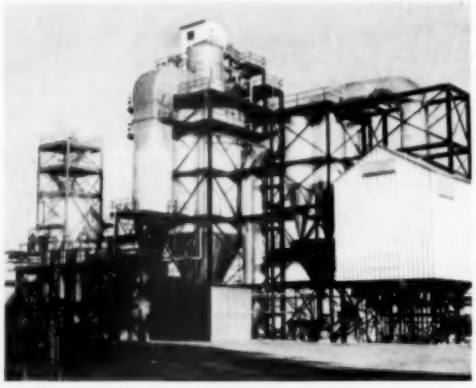

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extracting uranium, propelling rockets and maybe setting up space-stations, man is making a flippant bet with Nature that it will all come out right in the end. Meanwhile, let us—as people specially interested in food—note what the real situation is, and think what the consequences might be, say 40 years on.

Irrigation—often linked with supposedly "unlimited" energy expected from nuclear power—is an almost empty talking-point, ignoring economics as well as science; even if electricity cost nothing to generate, the costs of transmission would prevent its becoming much cheaper.

A rough estimate is that if the energy of three 400-megawatt power stations (the size of those now being built in England) were applied merely to pumping irrigation water from coast to desert, their liquid through-put might serve to nourish about one million people. That ignores the still vaster amount of energy needed to separate usable water from sea-water. Such notions for the future extension of irrigation might be truly called pipe-dreams. Evaporation of sea-water at present efficiencies requires burning of about half an inch of oil per acre to produce enough water to irrigate an acre, or say 40 tons of oil per annum to feed one or two people.

There is nothing in sight to justify the idea that *any* form of external physical energy—even sunlight—can give all of us now or in the future the human energy that can be obtained only by eating the right sorts of food.

Food is a substance; it cannot be generated by energy, but requires other substances for its production. The fact is that about a third of "Western" and Japanese people at present, and an increasing proportion of mankind in the near future, must depend for food on diminishing stocks of fuel under the surface of the earth.

Food science embraces all countries and all seas. It is very much a matter of soil and climate and therefore (one might suppose) of acquiring and disseminating knowledge over a wide area of land and thought, wherever a problem touching on food happens to exist—say of reclaiming alkaline soils or otherwise augmenting food-production in underdeveloped lands.

An archaeologist is not expected to do all his digging in his own back garden. If you pick up the *Radio Times* or daily newspaper, you will often find a talk or article by some architect, archaeologist, economist, or student of the outsides of birds who is acquainting the public with what he saw during a study-visit overseas. But you cannot use this yardstick with chemistry, because what chemists have to say is not usually supposed to interest the public.

A power of imperial rank might be expected to understand the need for extending food science abroad—the more so if its own income of food depends largely on imports of food. There seems to be no such awareness as to render exchange of ideas about food production easy, not even within the terri-

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tories of the Commonwealth; a British chemist whose study is food may as well accept the facts, first that he is confined to a region of these islands and, secondly, that his opportunities for giving formal teaching about fundamentals of food to the present and the coming generation, either at home or abroad, will be minimal or zero. The appropriate machinery does not exist.

The Fisheries Branch of the Ministry of Agriculture, Fisheries, and Food does have research ships—travelling laboratories—to take its scientists to the fish; nothing like that has been envisaged for the appreciation of man's relationships to his soils. It may be noted that the only recent organised exploration was of a continent where there is no human life, no soil and no plants.

The explanation of this neglect of study of the basic relations between man and food is, I think, to be found in education, and in the circumstance that impulses to acquire knowledge and found new techniques are conditioned by laymen; only those gaps about which laymen are conscious are likely to be filled without much effort in argument and persuasion by scientists—effort, that is, additional to scientific work proper. Harwell was not built by the process of subordinates petitioning for a few hundred pounds' worth of equipment at a time. Fundamental science cannot be prosecuted unless in a field of which laymen have grasped the importance, and it is safe to say that no layman has heard of chemical ecology—which is the chemical and fuel aspect of the relation of living things to the world.

Putting chemical ecology into plain language and calling it "food production for the future" does not yet seem likely to attract eager attention from those outside the Food Group. Most people will be unconcerned, being already comfortably convinced that there is really nothing to worry about—now that synthetic nitrogen, nuclear energy and a wheat ("food") surplus are with us and that international organisations have been set up to study food problems.

The truth is that existing agencies are tending to obscure major truths about the origins of food. To spread the facts, and to bring implications of established knowledge within reach of everybody, could be social service of high order. In a letter to *Chemistry & Industry* (February 15, 1958, p. 189) I suggested that there is an opportunity for the Society of Chemical Industry to take the leading place among counsellors about food and man. Education, like culture, needs to be a thousand men thick.

There is a long-standing failure of university chemistry departments to tackle fundamentals of food science. It was in 1858 that the chemist Louis Pasteur in Lille founded bacteriology, and Thomas Anderson, a medical man who was chemist to the Royal Highland and Agricultural Society and Regius Professor of Chemistry in Glasgow, introduced some pioneer chemical ideas about crops. Since about that time no fundamental or essentially novel idea about soil or crops or other non-homogeneous materials of biological importance has come from a university chemistry department, as far as I know.

Agricultural chemistry has too long had its fundamentals left to amateurs like J. B. Lawes and the late Sir George Wilson, and to sewage boards, agricultural colleges and other non-university bodies. In the last decades, brewery chemists alone have made more useful approaches to biological variables than a hundred years of university chemistry have done.

At the turn of this century, the contrast between the questing vigour of basic physics and the somnolence of food science was sharp. Young university men like Planck and Einstein were evolving ideas that form part of the mentality of thinking people today. When Lawes died, a very old man, in 1900, there was no tradition of food chemistry, and the spirit of enquiry was spent. There was, and is, no teaching effective enough to displace irresponsible notions like those of William Crookes about nitrogen: Lawes had stated the governing principle (p. 6), but it is still generally unknown. Seventy-odd years after a historical change, the tremendous fact that man himself is in an unprecedented food-relationship with his world awaits recognition.

It is true that food chemistry has never had a shake-up comparable to that administered by the discoveries of Becquerel and Röntgen. But nothing dramatic is needed. All that is required is an appreciation of ordinary basic chemistry and the ability to

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follow up the idea that food consists essentially of reduced carbon and nitrogen combined and obtained in particular ways.

The status of food science suffers from lack of attention to fundamentals. Though serious misunderstandings about food prevail among laymen, many scientists need reminders and correction about basic science. Thus: much that is written about "ions" (notably in Holland in reference to composition of herbage) does not apply to ions at all. Published "ionic ratios" include such constituents as protein-sulphur and total calcium. No inorganic chemist would refer to "the cobalt ion" in potassium cobaltinitrite!

That not only authors, but scientific editors and referees, see nothing odd in many aberrations of chemistry and logic in biological procedure does suggest how great is the need for a return to chemistry in work on food and nutrition and that more attention needs to be given to chemical ecology and so to the training of those who may be going to do ecological and biochemical work. Some biological branches of chemistry seem to have weakened themselves by becoming almost detached from the parent tree.

The bodies that have, or have had, fundamental interests in chemical ecology do either no teaching or little or none at graduate and post-graduate level. Is it surprising that there is intellectual stagnation about food supplies and the scientific principles by which food is produced?

So long as the universities are dismissive, the higher ranks that form opinion and allocate funds are unlikely to be penetrated by information about the origins of food. There is a serious risk that, in the second half of the twentieth century, public attention to food problems will continue to be held by discussion of the soundness of an eighteenth-century idea of Malthus as abutted by an echo from nineteenth-century wrong notions about photosynthesis and nitrogenous fertiliser. Has nothing really been learnt in food science since 1798, or, say, 1880? And can anyone imagine a dictum of Thomas Telford (who, like Malthus, died in 1834) being received now as a cardinal theme of engineering problems of the future?

If we must have an old idea, let us have a good one: from D. B. Reid's 1836 "Text-book":

". . . the juster views that are now entertained of the nature and objects of education, require the remodelling of a system which had its origin in times of comparative darkness, and when Chemistry was a very subordinate art."

Since we must all eat, and because the only form of energy that is able to keep us and our descendants alive is what is got from food having a due proportion of protein, these questions relate to social matters of interest to all scientists, having an impact on every citizen.

IN THIS ISSUE—

TECHNICAL ARTICLES IN BRIEF

Enzymatic spoilage *Brined cherries*

The softening of brined cherries by polygalacturonase, and the inhibition of polygalacturonase in model systems by alkyl aryl sulfonates. Wilbert F. Steele and H. Y. Yang.

The cherry brining industry in the Pacific Coast States has increased greatly since the passage of the Tariff Act of 1930. Until the passage of this act more than 80% of the brined cherries used for the manufacture of glace and maraschino cherries in the United States were imported from European sources. Since that time the percentage has been reversed until today most of the brined cherries used for this purpose in the United States are supplied domestically. There have been occasional occurrences of cherries softening in the brine. In this study no polygalacturonase activity could be demonstrated in firm ripe cherries, nor in cherries infected with cherry powdery mildew or brown rot. None of these appears to be involved in the softening of cherries during the brining and storage period. The alkyl aryl sulfonate detergent Nacconol NR was found to be an effective inhibitor for the polygalacturonase in Pectinol M in cherry brine at pH 2.6 and in buffer solutions at pH 3.0, 3.6 and 4.0. Inhibition of this enzyme by Nacconal NR at pH 4.0 was found to be directly proportional to the concentration of the detergent added for 0.1 and 0.2% Pectinol M solutions. This type of inhibition is typical of non-competitive enzyme inhibitors. The detergent Nacconol NR did not cause any deleterious effect when added to firm brined Bing or Royal Anne cherries at a concentration of 0.5% two weeks before processing into maraschino cherries. The effect caused by adding Nacconol NR to cherries at the time of brining was not determined and this should be done before it is used as a general inhibitor of polygalacturonase. The "cats claw" disease of Bing and Lambert cherries was found to contain a large concentration of a polygalacturonase, and these cherries caused rapid deterioration of brined cherries when 10% by count of the infected cherries were brined with firm Bing cherries. The cherries had deteriorated noticeably within 10 days after brining. Since the pectic materials are generally conceded to be responsible for the firm texture of brined cherries, it would appear that a polygalacturonase, rather than other types of polysaccharidases, is primarily responsible for the observed softening. The type of softening observed in these cherries was similar to that described by McCready and McComb in 1954, and to that found in commercially packed cherries from the 1957 season. This

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indicates that the type of spoilage which resulted in the softening of brined cherries during 1957 was probably identical to that caused by "cats claw" contaminated cherries. Little further deterioration was observed in these brined cherry samples after the enzyme activity had stopped. The period of breakdown was found to parallel the time of polygalacturonase activity. (See page 121.)

Antibiotics — *Preservation of harvested spinach*

Factors affecting decay of prepackaged spinach. R. F. Becker, R. N. Goodman, and H. S. Goldberg.

To overcome the harmful effects of high temperature, refrigeration has been widely used to reduce the incidence of soft rot in prepackaged spinach. In commercial practice, excess wash water is commonly removed by centrifuging the spinach prior to bagging. However, despite refrigeration, centrifugation, and careful handling, the shelf life of prepackaged spinach is often limited to a very few days. Previous work showed that, under conditions of rapid decay, 25 meg/ml solutions of oxytetracycline, chlortetracycline and streptomycin sulfate retarded the development of soft rot and reduced the population of bacteria in prepackaged spinach. Data from the present study indicate that spinach grown during the hot summer months is more susceptible to decay than that grown at other times of the year. Experimental evidence has shown that total bacterial population and the population of actual soft rot bacteria are directly proportional to rate of decay. There is no such relationship in the case of yeast and fungi. Thus, rate of decay appears to depend on the bacterial population of the spinach. However, other factors, such as the nitrogen content of the leaves, may also play a part in determining the decay rate, either by affecting the microorganisms or by predisposing the spinach to attack. Evidence obtained from this study suggests that antibiotics used at low concentrations have practical applications in the preservation of leafy vegetables such as spinach. It is felt that these chemicals would be particularly effective in reducing the number of bacteria during warm weather, when soft rot development is most rapid. To date, the Federal Drug Administration has not approved the use of antibiotics in the preservation of harvested vegetables. However, since spinach is usually eaten after it is cooked, heat labile antibiotics such as oxytetracycline and chlortetracyline may in the future find a place in the preservation of harvested leafy vegetables. (See page 127.)

Packaging — *Collapsible polyethylene tubes for food*

The collapsible polyethylene tube as a food package. J. D. Kemp, R. M. Ballantyne, A. J. Ducker, and J. W. Haynes.

Many types of military field rations contain spreads such as jam, jelly and peanut butter. Individual rations require a light weight package containing about two ounces of such a spread. Standard commercial packages are usually cans or glass jars which contain quantities sufficient for several meals. The smallest sanitary can commercially available in Canada holds four ounces of jam and the only individual portion package—the small, formed plastic cup used by restaurants and airlines—is too fragile for military use. To satisfy the need for a suitable package—one that is light in weight, will protect the food satisfactorily for two years at 70° F, will withstand a moderate degree of rough usage, is capable of being reclosed, and will not present a serious damage hazard to other packages

with which it might be assembled—the use of collapsible polyethylene tubes was considered. The tubes proved to have considerable strength. A filled and properly sealed tube laid on its side would support one hundred and fifty pounds weight without rupturing; therefore the tubes were not regarded as being a serious breakage risk in field rations. A series of storage experiments was conducted in the cold (0° F), at a normal room temperature (70° F) and at an elevated temperature (100° F). The products were raspberry jam, grape jelly, honey, peanut butter and creamery butter. Lined (i.e., coated with polyvinylidene chloride resin) and unlined collapsible polyethylene tubes were the containers. Lined tubes were found to be more suitable than unlined tubes because of a lesser tendency to produce off-flavours and because of their impermeability to oils. This impermeability indicated their potential suitability as packages for oily foods such as butter and peanut butter, but oxygen transmission characteristics of the tubes should be investigated more fully. During comparative tests at 100° F the storage life of jam, jelly and honey in lined tubes was generally comparable to that in bottles or cans for periods up to three months; thereafter, the quality of foods in bottles and cans was slightly superior. (See page 131.)

Method to determine doneness — *Defrosted turkey halves*

Doneness of frozen, defrosted turkey halves roasted to several end point temperatures. Graycee S. Goertz, Katherine Cooley, Martine C. Ferguson, and Dorothy L. Harrison.

Before a reliable and uniform research method to roast turkey can be formulated, various procedures for determining end points need further examination. The purpose of this study was to investigate certain end point temperatures in both breast and thigh to determine which would produce most consistently an optimumly done bird. Eighty frozen defrosted Broad Breasted bronze turkey halves were roasted at 163° C (325° F) to end point temperatures of 85° and 90° C in the *pectoralis* major and 85°, 90° and 95° C in the thigh. Turkey halves roasted to either 90° C in the *pectoralis major* or 95° C in the thigh were satisfactorily done. However, an end point temperature of 90° C in the *pectoralis major* was preferred to 95° C in the thigh because of the size and greater uniformity of the *pectoralis major* muscle. Turkeys roasted to 85° C in the *pectoralis major* and 90° C in the thigh were similar and less done than those roasted to 90° C in the *pectoralis major* or 95° C in the thigh. Turkey halves cooked to end point temperatures of 85° C in the thigh were underdone. Flavor and tenderness scores were similar for birds cooked to all end point temperatures. Juiciness scores and press fluid yields were lower and cooking losses higher for satisfactorily done than for underdone turkey halves. The amount of heat-coagulable protein in the *pectoralis major* was not related to the doneness of the muscle. (See page 135.)

Evaluation methods — *Darkening of dates*

Studies on domestic dates. I. Methods for evaluating darkening. V. P. Maier and Frenk H. Schiller.

(Continued on page 24 following Technical Section)

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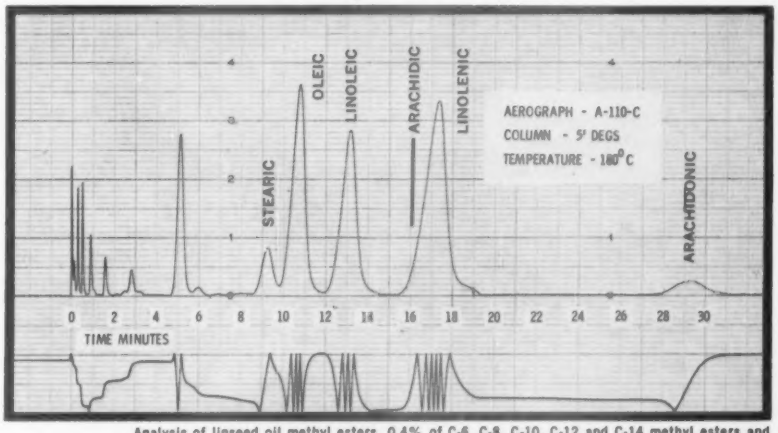

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(Continued from page 21 preceding Technical Section)

Studies on the chemistry of deterioration of domestic dates necessitate the development of reliable objective methods for measuring their darkness. Such methods make possible the quantitative determination of the effects of variables such as temperature, moisture content and oxygen concentration on the rate of darkening. Objectives of the work reported in this paper therefore were (1) to develop a reflectance method for measuring the darkness of dates; (2) to develop a solvent extraction method for measuring the amount of dark pigment produced in darkened dates; and (3) to evaluate and compare both methods over a wide range of darkness values using identical samples of dates. Two methods have been developed by which the darkening of dates during deterioration can be followed. The reflectance method in which either whole or pureed dates can be used is very sensitive to the initial darkening and is closely related to the visually observed darkness. Use of pureed dates is preferred, however, because of the greater precision and the broader range of reflectance values obtained with light and dark dates. The solvent extraction method permits the precise measurement of the amount of soluble dark pigment present in date tissue of any degree of darkness. Although the results are not directly related to the visually observed darkness, this method has the advantage of providing a measure of the later stages of darkening where the reflectance method is not sensitive. (See page 139.)

Antibiotics and irradiation *Round steaks*

The use of antibiotics and gamma irradiation in the aging of steaks at high temperatures. G. D. Wilson, P. D. Brown, W. R. Chesbro, Betty Ginger, and C. Edith Weir.

Tetracycline antibiotics alone and in combination with gamma radiation were used as preservatives during aging at elevated temperatures. It has been reported that high energy irradiation and the tetracycline antibiotics complement each other in their preservative effect on meat. The adverse effects of irradiation on meat flavor are generally proportional to the dosage empoyed but are not pronounced at "pasteurization" levels. Beef has been shown to be more susceptible than pork to flavor and odor alteration. In the present study, both antibiotics and irradiation were used, when needed, to allow maximum flexibility in holding conditions with the objective of establishing the optimum aging time and temperature for the rapid tenderization of beef. Round steaks from U. S. Utility and U. S. Good carcasses were used to determine the optimum conditions of high temperature and time for increasing the tenderness of beef by aging. Microbial growth during aging was effectively controlled by infusion of 30–50 ppm of oxytetracycline into the rounds prior to cutting, or by 45,500 rad of gamma radiation in combination with the antibiotic treatment. At the level used (45,500 rad), gamma irradiation did not affect the tenderness or tenderization of steaks aged at 110° F for 24 hours. In other experiments, 2 days at 90° F and 24 hours at 110° F were found to be optimal for aging steaks to achieve effective tenderness. Steaks aged for 24 hours at 120° F showed greater increases in tenderness than those aged for 24 hours at 110° F or for 2 days at 100° F. Aging at 90° F necessitated more severe treatment to provide excessive bacterial growth; aging at 120° F resulted in more undesirable color and flavor changes in the steaks than found in those aged at the lower temperatures. On this basis, 24 hours at 110° F was selected as the most desirable aging time and temperature for the rapid tenderization of the beef round steaks. Additional studies showed that steaks aged for 24 hours at 110° F had tenderness increases comparable to those found in steaks aged for 14 days at 35° F. (See page 143.)

Freezing technology *Boysenberries*

Time-temperature tolerance of frozen foods. XX. Boysenberries. D. G. Guadagni, K. M. Eremia, S. H. Kelly, and Jean Harris.

Detailed results on time-temperature tolerance of major frozen fruits in retail packages have been reported previously. This paper gives results on effect of temperature in larger than retail packages. Since frozen boysenberries are mainly prepared in individually quick frozen form and packaged in institutional sizes for remanufacture into pies and other products, the principal effort in the experiments reported was devoted to this type of product and size of package. Results on moisture, ascorbic acid, color, and flavor changes in both individually quick frozen and sirup-packed frozen boysenberries held at temperatures from 0° to 30° F are reported. Stability of siruped boysenberries in retail packages was very good in the temperature range of 0° to ±10° F, but individually quick frozen boysenberries loosely packed in cardboard cartons were somewhat less stable at the same temperatures. At 20° to 30° F the apparent stability was in favor of the individually quick frozen fruit. Color diffusion from berries to sirup provides a means of estimating temperature history in the retail packs. Time required for accumulation of 10 to 12 mg/100 g of oxidized ascorbic acid (DHA + DKA) corresponded to the approximate high-quality life (as determined by panel measurements) of both siruped and individually quick frozen berries held at temperatures of 10° to 30° F. Moisture loss in individually quick frozen berries detracted from appearance of the top layers of fruit and reduced the package net weight by 6 to 8% during the high-quality life of samples held at 0° to 10° F. At 20° F, definite quality changes occurred when only 2 to 3% weight loss was recorded. (See page 148.)

New product (potential) *Cherry seed oil*

Composition of cherry seed and cherry seed oil. (Prunus cerasus). K. G. Weckel and H. D. Lee.

The processing of cherries results in the disposal of very large quantities of cherry seeds. In Wisconsin and Michigan, the total cherry production averages 120 million pounds annually. The seeds are not used to provide income to the canners or growers. Kernels from cherry pits have been reported to contain an oil resembling that of almond. The amount of oil in the kernel ranges from about 30 to 38% of the dry kernel weight. The weight of the kernel represents 28% of the total cherry pit weight. The moist cherry pit accounts for about 10% of the total cherry weight. The weight of the moist cherry pit is about twice the weight of the dried cherry pit. From these data, it has been estimated the amount of cherry kernel oil potentially available in Wisconsin and Michigan is 100,000 gallons. Cherry kernels have a fairly high protein and starch content which may have potential in animal and food mixtures. The composition of cherry kernel oil resembles that of almond oil and is quite similar to that of corn oil, peanut oil and sesame seed oil. Elaeostearic acid was shown to be present in cherry kernel oil. The presence of elaeostearic acid may be the cause of accelerated development of oxidative rancidity observed in the oil. (See page 151.)

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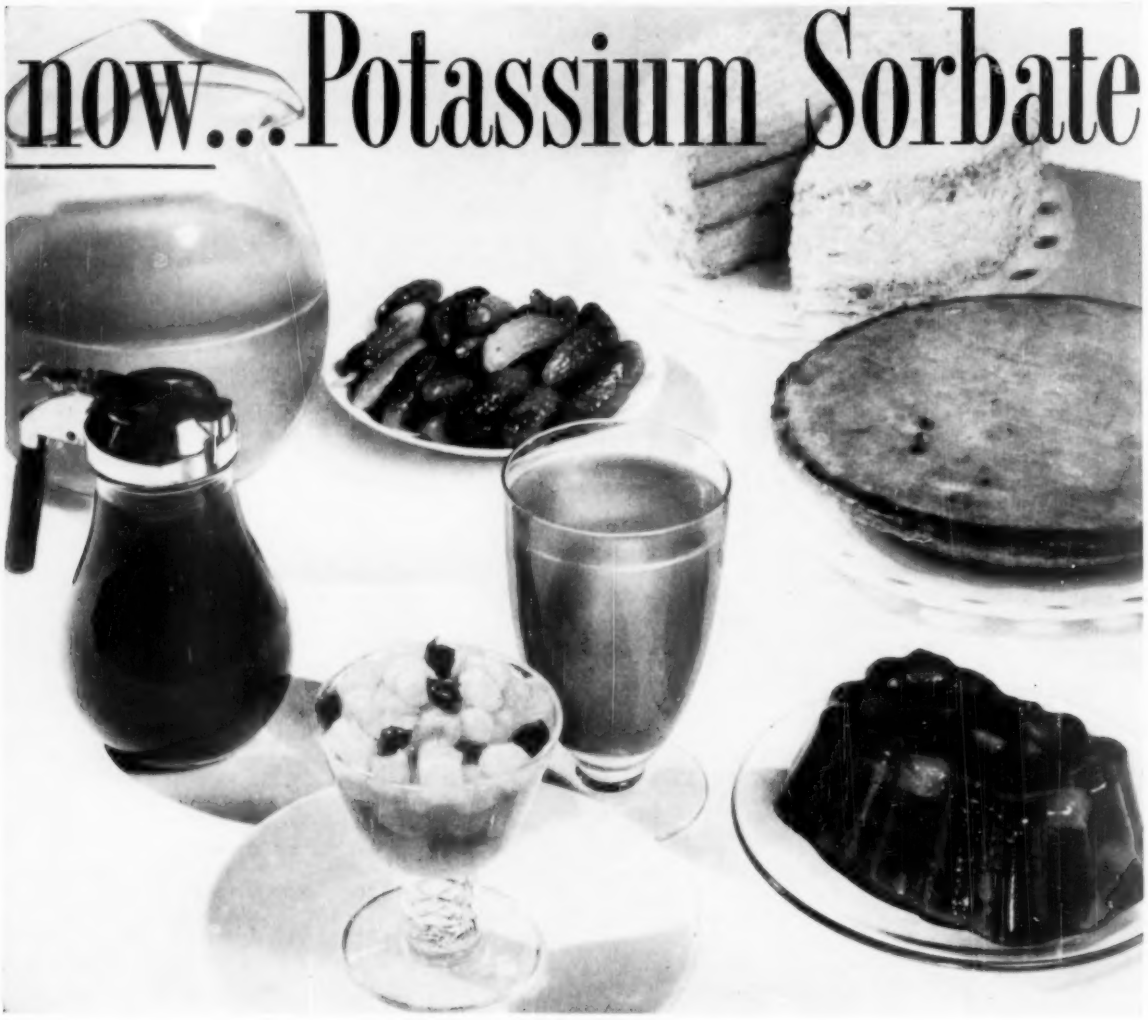

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The Softening of Brined Cherries by Polygalacturonase, and the Inhibition of Polygalacturonase in Model Systems by Alkyl Aryl Sulfonates[a, b]

Wilbert F. Steele and H. Y. Yang

Department of Food and Dairy Technology, Oregon State College, Corvallis, Oregon

(Manuscript received September 23, 1959)

THE CHERRY BRINING INDUSTRY in the Pacific Coast States has increased greatly since the passage of the Tariff Act of 1930. Until the passage of this act more than 80% of the brined cherries used for the manufacture of glace and maraschino cherries in the United States were imported from European sources. Since that time the percentage has been reversed until today most of the brined cherries used for this purpose in the United States are supplied domestically.

The brining method which has proved to be most suitable for the preservation of cherries in the Pacific Northwest was developed by Bullis and Wiegand in 1931 *(2)*. This procedure employs a brine containing approximately 1.25% sulfur dioxide to bleach the cherries and to prevent microbial growth, and 0.7% calcium hydroxide—or its equivalent in other calcium salts—to form a firm gel with the pectic materials present in the cherries. This has proved to be generally satisfactory, but there have been occasional occurrences of cherries softening in the brine. This type of damage was first noticed in the Yakima, Washington area about 1947 *(24)*.

McCready and McComb *(16)*, in 1954, published the first report on this type of spoilage of brined cherries. They observed that, " . . . soft brined cherries seem to be attacked from the outside by a pectin degrading enzyme, since the stems fall off and part of the outer skin begin to slough. The pectic substances are slowly attacked during the brining storage period and then are so badly degraded that they are no longer capable of forming firm calcium pectate gels within the cherries."

Since the damage was scattered and slight, little further investigation was made of the cause or source of the spoilage agent. During 1957, however, extensive damage to brined cherries occurred in Oregon and Washington. This softening, as in the previously reported cases, was not noticed until the crop was being prepared for shipment to processors, which was several weeks after the crop had been brined. Consequently, the cherry briners affected suffered a heavy loss, estimated at more than 11,800 barrels valued at approximately $516,000 *(24)*.

The softening observed during 1957 appeared to be identical to that reported by McCready and McComb *(16)*. However, the cause was still not known and it was necessary to set up a general program to (a) determine the source and nature of the agent responsible for softening, and (b) search for a method of preventing damage to the cherries after brining.

From the nature of the softening as related to the brining procedure it appeared that a pectin degrading polygalacturonase was responsible since this enzyme is capable of hydrolyzing the 1,4-glycosidic linkages of pectinic and pectic acids. Such an enzyme might result from microbial growth on the fruit prior to brining, then act on the fruit during the brine curing period. Two other possible sources of polygalacturonase were the fruit itself which might synthesize the enzyme during ripening, or certain cherry diseases. The disease cherry powdery mildew (*Podosphaera oxyacathae* D. C. de By), which was prevalent during 1957, was one of these.

Literature background. The report of McCready and McComb *(16)* is the first in which the enzymatic softening of brined cherries is indicated. Investigators at the U.S.D.A., Western Utilization Research and Development Division *(25)*, Ross, Yang and Brekke *(23)* and Wiegand *(24)* are in agreement that softening of brined cherries results from the action of pectin degrading enzymes.

The softening of cucumbers in commercial and model fermentations has been studied by Etchells, Bell and Jones *(5)* who have found pectic enzymes in cucumber flowers which contained a number of fungi (Fusarium, *Penicillium*, *Alternaria*, *Mucor*, and *Aspergillus*). The inhibition of this enzymatic action in cucumber fermentations by scuppernong grape leaves has been reported by Etchells, Bell and Williams *(6)*.

The softening of strawberries and other small fruit preserved in sulfurous acid solutions without heat treatment has occurred in England, according to Joslyn and Braverman *(8)*. This has been attributed to the presence of enzymes secreted by the microorganisms (chiefly molds) contaminating the fruit, and not to the activity of naturally occurring enzymes present in the fruit.

The occurrence of polygalacturonase activity in ripe pears and avocado has been reported by McCready and McComb *(17)*. They did not detect any polygalacturonase activity in unripe pears, peaches, or avocado.

The action of pectic enzymes has been reviewed by

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[a] Approved for publication by the Director of the Oregon Agricultural Experiment Station as Technical Paper No. 1264. Contribution of the Department of Food and Dairy Technology.

[b] Partly based on a thesis by Wilbert F. Steele, submitted in partial fulfillment of the Master of Science Degree.

Kertesz (*10*, p. 333–375), who concluded that there was sufficient evidence to justify the naming of a pectin-methylesterase, which hydrolyzes the methyl ester group of pectinic acids, and polygalacturonase, which is capable of hydrolyzing the 1,4-glycosidic linkages of pectinic and pectic acids. McCready, McComb and Jansen (*19*) found no difference in the reaction of polygalacturonases from avocados, tomatoes, or fungi.

Pectin-methylesterase occurs in the roots, stems and leaves of many higher plants. Phaff and Joslyn (*21*) report that it is produced by many fungi and bacteria. Demain and Phaff (*3*), Kertesz (*10*), Lineweaver and Ballou (*13*) and MacDonnell, Jansen and Lineweaver (*14*) have reported that this enzyme is capable of attacking methyl esters of pectinic acid with the formation of pectic acid and methanol. Pectin-methylesterase activity in the presence of polyvalent ions—especially calcium salts—results in the formation of a firm calcium pectate gel (*10*) but no gel is formed in the absence of metallic ions. Phaff and Joslyn (*21*) report that pectin-methylesterase is often found associated with pectic enzyme complexes.

The action of pectin polygalacturonase has been studied extensively. The hypothetical degradation by hydrolysis of the 1,4-glycosidic linkages of pectinic and pectic acids has been supported by the work of Phaff and Demain (*20*), who have isolated a single polygalacturonase from yeast as shown by electrophoresis and by ultracentrifuge studies. These authors observed a rapid, random, initial hydrolysis of pectic acid with the production of tetra-, tri-, di-, and galacturonic acids. The rate of hydrolysis was found to decrease as the reaction proceeded.

McCready and Seegmiller (*18*) found that purified polygalacturonase attacks polygalacturonide substrates which possess a glycosidic uronide bond between two adjacent free carboxyl groups, but fails to attack those where one or both of the adjacent carboxyl groups is esterified. Pectin-methylesterase has been found to increase the activity of polygalacturonase when present in moderate concentrations (*21*).

The inactivation of pectin-methylesterase by the alkyl aryl sulfonate detergent Nacconol[c] has been reported by McColloch and Kertesz (*15*). This detergent was reported to be an effective inhibitor of the pectin-methylesterase of tomatoes, but was found to be less effective in inihibiting pectin-methylesterase of fungal origin. Experiments with other detergents and soaps gave varying results. None of the common enzyme inhibitors, such as heavy metals, had any inhibiting effect on this enzyme. Since complete inhibition of the enzyme was achieved above a certain minimal concentration of the Nacconol detergent, they concluded that inhibition was due to surface denaturation of the protein, rather than to a combination of the detergent with reactive groups of the enzyme. Anson (*1*) has shown that this detergent will cause the denaturation of the proteins of hemoglobin and egg albumin at the isoelectric point.

Kertesz (*10*) has reported that Nacconol is much less effective as an inhibitor of polygalacturonase. Rahman and Joslyn (*22*) experimented with the inhibition of the polygalacturonase of purified Pectinol[d] fungal preparations by use of mercurous chloride, sodium fluoride, iodoacetic acid and sulfur dioxide with no apparent inhibition occurring. In experiments on the pH optimum they reported optimum activity at pH 4 to 5, a very low activity at pH 3.0 and decreasing activity from pH 5 to zero activity at pH 8.0.

Studies on the calcium treatment of apples and tomatoes by Laconti and Kertesz (*11*) and Kertesz (*9*) have shown that firm calcium-pectin gels are formed by the reaction of calcium with low ester pectic substances to form insoluble calcium pectates and calcium pectinates. Demain and Phaff (*3*), in their review of the softening of cucumbers, conclude that, " . . . apparently spoilage can occur via the conversion of insoluble pectic material (protopectin) to more soluble forms and smaller molecules without the loss of calcium pectate forming ability."

EXPERIMENTAL PROCEDURE

I. Study of Fresh Cherries

A. Measurement of polygalacturonase activity. Fresh sweet cherries of the major brining varieties—Bing, Lambert, Royal Anne, Black Republican—were tested for polygalacturonase activity by a modified procedure based on the "cup plate" technique developed by Dingle, Reid and Solomons (*4*).

A polygalacturonic acid substrate medium was prepared by slowly suspending 0.25% (w/v) of polygalacturonic acid (Exchange Lemon Products Co., No. 491) in distilled water with constant stirring. Next, 0.25% ammonium oxalate, 1% potassium biphthalate and 0.1% thymol preservative (all w/v) were added and dissolved. The pH was adjusted to 4.0 with 0.1N sodium hydroxide. The medium was transferred to an Erlenmeyer flask and 2% bacto agar (Difco) added, and sterilized by autoclaving for 15 min at 15 psig. Then 25 ml aliquots were transferred to 15 × 150 mm Petri dishes and allowed to solidify. Excess medium was stored in stoppered bottles until needed.

A standard curve was prepared representing the polygalacturonase activity of Pectinol M solutions in distilled water at concentrations from 0.005 to 1.0% by serial dilution of a stock 1.00% solution (w/v of the air dried unpurified preparation). For each test, one drop (approximately 0.05 ml) of the test solution was placed on the flat surface of the test plate, then the plate was incubated for 20 hr at 37° C, and developed by flooding with 2 N hydrochloric acid for 5 min and then rinsing with distilled water. Polygalacturonase activity was indicated by formation of a clear zone, and the extent of this activity could be estimated by measuring the diameter of the zone formed. The average diameter of triplicate determinations was used for the establishment of a reference curve.

The standard curve obtained by plotting the diameter of clear zone formed against the log per cent of the concentration of Pectinol M gave a straight line over a wide range of concentration. These data for the Pectinol M solutions by the drop method used are given in Table 1.

A comparison was made of results obtained by use of the "cup plate" method of Dingle, Reid and Solomons (*4*), and a "drop" method where the test solution was placed directly on the flat surface of the substrate without cutting cups.

[c] Manufactured by National Aniline Division, Allied Chemical and Dye Corp., New York, N. Y.

[d] Pectinol is the trade name for the pectic enzyme preparations manufactured by Rohm and Haas Company, Philadelphia, Pennsylvania.

TABLE 1

Diameter of clear zone as a function of concentration of Pectinol M. Solutions

Concentration, per cent (w/v)	Diameter of clear zone formed (mm)[1]
0.005	x
0.01	8.0
0.05	15.5
0.1	18.0
0.5	20.5
1.0	22.0

x Not measurable.

[1] All plates contained one drop (approximately 0.05 ml) per test. Results are averages of triplicate determinations. Incubation period was 20 hours at 37° C.

Equivalent results were obtained by either method, but because of the difficulty encountered in cutting and removing cores to obtain approximately circular cups, and because of the further difficulty involved in trying to place and measure the test solution properly in these cups, the "cup plate" method was discarded for the simpler "drop" method.

B. Determination of polygalacturonase activity in fresh cherries. Fresh cherries of several varieties were obtained during the 1958 season and tested for polygalacturonase activity. For these tests the fruit was macerated with a mortar and pestle and one drop of the liquid (approximately 0.05 ml) was placed on the surface of the polygalacturonic acid substrate with the aid of a 1 ml pipette, and the plates incubated and developed as previously described. Although many of these cherry samples were ripe and highly colored, this coloration did not interfere with the determination.

Cherries infected with brown rot, cherry powdery mildew and "cats claw" were also tested during this period. Polygalacturonase activity could be demonstrated only in ripe Bing and Lambert cherry varieties infected with what has been termed "cats claw" damage. This spoilage is characterized by claw-like markings on an area of approximately one-fourth of the cherry, and this portion of the fruit has a soft and mushy appearance with a considerable exudation of juice, although the remaining portion of the cherry is firm and sound with no apparent defect.

Microorganisms from "cats claw" infected cherries were isolated and cultured on nutrient agar slant cultures, then transferred to a polygalacturonic acid-nutrient agar substrate-medium which contained all the ingredients of these two media except the thymol preservative. After inoculation, the plates were incubated at 27° C until growth occurred, then developed as previously described. The secretion of a polygalacturonase during growth of an organism was demonstrated, when it occurred, by a clear zone at the site of growth. No quantitative estimation of this activity was attempted because of the variations in growth patterns and incubation periods of the microorganisms selected.

Isolated microorganisms which produced a polygalacturonase during growth were identified and classified by the usual procedures.

Samples of "cats claw" damaged Bing cherries were brined in a standard 1.25% sulfur dioxide-calcium hydroxide (0.7%) cherry brine by placing 10% by count of the damaged fruit with 90% of firm ripe Bing cherries. These samples were tested periodically for polygalacturonase activity and observed for softening.

II. Studies on the Inhibition of Polygalacturonase

The inhibiting action of the commercial alkyl aryl sulfonate detergent Nacconol NR was determined quantitatively after preliminary studies had shown it to be an effective inhibitor for the polygalacturonase of Pectinol M. For these studies a 5.0% (w/v) solution of Nacconol NR was prepared from the air dried powder (40% active ingredients). This stock solution was used to add the required amount of detergent to clean dry 200 ml capacity screw cap bottles from a 10 ml microburette. Then 50 ml of freshly prepared 0.1 and 0.2% (w/v) Pectinol M solution in pH 4.0 Clark and Lubs buffer was added at each concentration of the detergent, and the flasks shaken for 30 sec to insure mixing. An aliquot then was withdrawn and tested for polygalacturonase activity. Approximately 0.05 ml was used per test, but the quantity had to be estimated because of the large decrease in surface tension caused by the addition of Nacconol NR. Tests were conducted at intervals of ¼, ½, 1, 2, 3, 24 and 48 hr. The rate of inhibition of enzyme activity was measured as per cent of the maximum activity observed in control samples which contained no added Nacconol NR.

The effect of adding Nacconol NR to cured firm brined Bing and Royal Anne variety cherries was observed by adding 0.1, 0.3, and 0.5% (w/v) of the detergent to these cherries. The cherries were left in contact with the detergent for 2 weeks, then processed into maraschino cherries by the method suggested by Filz and Henney *(7)*, and compared with control samples. No difference was observed in the treated or untreated samples.

RESULTS

I. The Polygalacturonase Activity of Fresh Cherries

Results of the polygalacturonase tests of fresh cherries are given in Table 2. No enzyme activity was found in cherries infected with brown rot, cherry powdery mildew (*Podosphaera oxyacanthae* D. C. de By), or in unripe cherries. A slight clear zone formed with all ripe firm cherries tested but this was too diffuse to measure and was considered to represent the normal occurrence of polygalacturonase in the mature fruit.

TABLE 2

The polygalacturonase activity of fresh cherries

Description of fruit and disease	Polygalacturonase test on macerated fruit pulp
Lambert, severe brown rot	negative
Bing, "cats claw" (whole fruit)	positive, 23 mm diam. clear zone
Bing, "cats claw" (damaged portion) of fruit	positive, 23 mm diam. clear zone
Bing, "cats claw" (undamaged portion of fruit)	negative
Lambert, "cats claw"	positive, 21 mm diam. clear zone
Cherry powdery mildew (*Podosphaera oxyacanthae* D. C. de By)	negative
Firm sound fruit—all varieties tested (ripe and unripe)	negative

All "cats claw" damaged cherries tested contained a large amount of polygalacturonase activity. This cherry spoilage, as was mentioned previously, does not cover the total surface of the cherry. When the firm, undamaged, portion of these cherry samples was tested, no polygalacturonase could be demonstrated. The remaining infected portion of the cherry contained all of the enzyme activity.

II. Tests of "Cats Claw" Infected Bing Cherries

A. Observations of brined cherries. Brined cherry samples prepared from "cats claw" infected fruit were tested periodically for polygalacturonase activity. Samples obtained from two different growers (A and B) were prepared. Results of the enzyme tests with these 2 samples are given in Table 3. The polygalacturonase activity decreased rapidly in these samples and had stopped completely in sample A after 13 days. Polygalacturonase activity was found in sample B at 13 days, but had stopped by the 25th day.

TABLE 3

The polygalacturonase activity in brined "cats claw" infected Bing cherries

Sample code	Per cent polygalacturonase activity[1]			
	Days in brine			
	2	6	13	25
A	0.05	0.03	0[2]	0
B	0.15	0.08	0.05[2]	0

[1] Expressed as % (w/v) of polygalacturonase activity found in Pectinol M stock solutions.

[2] Softening and breakdown of brined cherries was very extensive.

Softening and disintegration of these samples was noticeable after 10 days and had become extensive in both samples after 13 days. Sample B deteriorated to a greater extent, but both samples became soft and mushy; tht pulp separated from the pits to a great extent and the stems became separated from the pulp, or were easily detached in both samples. The appearance of these cherries during and after softening was similiar to that reported by McCready and McComb in 1954 *(16)*. Little change was observed in these samples once the polygalacturonase activity had stopped. This is similar to observations made on samples of commercial soft cherries obtained from the 1957 pack which have been kept in the laboratory for over a year with no signs of further degradation. The appearance of brined cherries degraded from "cats claw" infection and of commercially packed 1957 soft brined cherries was quite similar, indicating that the softening of cherries in both cases probably resulted from an identical or very similar cause.

B. The polygalacturonase activity of microorganisms. Several microorganisms were isolated from "cats claw" infected cherries, and found to produce a polygalacturonase under the test conditions used, and were identified as: *Rhizopus nigricans*, *Aspergillus niger*, *Saccharomyces niger*, *Penicillium*, *Rhodotorula* and apiculate yeast. Several microorganisms were isolated which did not produce this enzyme under these conditions. No attempt was made to identify these latter organisms, but most were yeasts.

These results show that there are several microorganisms present in this type of infection which are capable of producing a polygalacturonase. However, no mold growth was observed on the damaged cherry samples tested. Since the infected cherries were at an advanced stage of infection, when obtained, it would seem that if the polygalacturonase is produced by microbial growth, it is produced primarily by yeasts.

Since it is not likely that any microbial growth occurs after the cherries have been brined, the total pectic enzyme must be produced before brining. The polygalacturonase of infected cherries remained active in the sulfur dioxide brine, and occurred in sufficient concentration to cause rapid disintegration of the majority of the fruit present, although only 10% of the original fruit was contaminated.

III. Inhibitors of Polygalacturonase

Preliminary tests demonstrated that Nacconol NR would cause complete and apparently irreversible polygalacturonase inhibition when used in low concentrations. Quantitative estination of the degree of inhibition was then carried out. These data are shown in Tables 4a and 4b. Complete and immediate inhibition of the polygalacturonase in 0.1% Pectinol M solutions was achieved at Nacconol NR concentrations of 0.024% and greater in pH 4.0 buffer. Complete polygalacturonase inhibition of 0.2% Pectitnol M solutions in pH 4.0 buffer, was obtained at a Nacconol NR concentration of 0.04%.

A graph of the per cent of maximum polygalacturonase activity versus the per cent of Nacconol NR inhibitor added showed a straight line relationship to exist for the 0.1 and 0.2% Pectinol M solutions at zero time. This type of relationship is typical of that for non-competitive enzyme inhibition, where the inhibitor combines with some part of the enzyme essential for catalytic action. Surfac denaturation of the enzyme protein, as suggested by McColloch and Kertesz *(15)* for the inhibition of the pectin-methylesterase of tomatoes, may well be the cause of this inhibition. Anson *(1)* has shown that Nacconol causes denaturation of the proteins of hemoglobin and egg albumin.

Table 5 shows the inhibition of the polygalacturonase of 0.1% Pectinol M solutions at pH 3.0, 3.6, and 4.0. The enzyme inhibition was found to vary slightly with pH, although the polygalacturonase was inhibited completely at all pH values by a sufficient concentration of the Nacconol inhibitor. Studies on the pH optimum of this enzyme had shown maximum activity in the range of 4 to 5 for the polygalacturonase of Pectinol M.

The inhibition of the polygalacturonase of 0.1% Pectinol M solutions by Nacconol NR in a standard calcium hydroxide-sulfur dioxide cherry brine at pH 2.6 was determined and found not to differ greatly from the results obtained with buffer solutions. Complete and immediate inhibition of the enzyme was obtained at Nacconol NR concentrations of 0.03% and greater.

The surface active agents Igepal DJ-970, Span 20, Tween 40 and Tween 81 were tried as inhibitors of the polygalacturonase of Pectinol M solutions and found to have no inhibiting effect. Slow inhibition of this enzyme was obtained by the use of the household detergents Joy and Tide. This inhibition might be due to the presence of alkyl aryl sulfonates in these two all-purpose detergents since this type of detergent often contains more than one surface active agent.

TABLE 4

The inhibition of Pectinol M polygalacturonase by Nacconol NR

a. Inhibition in 0.1% solutions at pH 4.0.

Per cent Nacconol (w/v)	Per cent of maximum polygalacturonase activity[1] after the indicated time (hours)							
	0	¼	½	1	2	3	24	48
0	100		100					95.0
0.002	90.0	90.0	90.0	85.0	70.0	50.0	25.0	0
0.004	80.0	70.0	70.0	65.0	65.0	40.0	0	0
0.008	65.0	25.0	25.0	0	0	0	0	0
0.012	55.0	0	0	0	0	0	0	0
0.016	45.0	0	0	0	0	0	0	0
0.020	0	0	0	0	0	0	0	0
0.024 to 0.040	0	0	0	0	0	0	0	0

[1] Maximum circle diameter was 18.0 mm at zero time in control samples.

b. Inhibition in 0.2% solutions at pH 4.0.

Per cent Nacconol (w/v)	Per cent of maximum polygalacturonase activity[1] after the indicated time (hours)							
	0	¼	½	1	2	3	24	48
0	100		100			100	100	
0.002	95.0	90.0	90.0	85.0	80.0	75.0	45.0	30.0
0.004	90.0	80.0	80.0	65.0	60.0	50.0	30.0	0
0.008	80.0	70.0	50.0	40.0	0	30.0	0	0
0.012	70.0	40.0	30.0	25.0	0	0	0	0
0.016	55.0	0	0	0	0	0	0	0
0.020	45.0	0	0	0	0	0	0	0
0.024	40.0	0	0	0	0	0	0	0
0.028	30.0	0	0	0	0	0	0	0
0.032	20.0	0	0	0	0	0	0	0
0.040	0	0	0	0	0	0	0	0

[1] Maximum circle diameter was 19.5 mm at zero time in control sample.

TABLE 5
The inhibition of Pectinol M polygalacturonase by Nacconol NR at pH 3.0, 3.6 and 4.0

pH	Per cent Nacconol NR (w/v)	Per cent of maximum polygalacturonase activity[1] after the indicated time (hours)				
		0	½	1	24	48
3.0	0	100				95.0
3.0	0.01	80.0	0	0	0	0
3.0	0.03	0	0	0	0	0
3.0	0.05	0	0	0	0	0
3.6	0	100				100
3.6	0.01	90.0	70.0	55.0	0	0
3.6	0.03	60.0	0	0	0	0
3.6	0.05	0	0	0	0	0
4.0	0	100				100
4.0	0.01	85.0	80.0	10.0	0	0
4.0	0.03	70.0	0	0	0	0

[1] Per cent of maximum activity of control sample at each pH value immediately after preparation. Pectinol M solutions were 0.1% (w/v).

DISCUSSION

A polygalacturonase was found to occur in excessive amounts only in fresh cherries infected with "cats claw" type of spoilage. These same cherries underwent rapid and extensive deterioration when brined, and the polygalacturonase was found to remain active for a period of from 13 to 25 days in the brined cherries. Once the enzyme activity had stopped, no further change was noted in the appearance of the cherries, even after an additional seven month storage period.

Since the appearance of these soft cherries was quite similar to that of soft commercially packed cherries obtained during the 1957 season, the "cats claw" type of spoilage in fresh cherries is probably responsible for the occurrence of cherry breakdown in commercial packs. The fact that this enzyme is capable of attacking the cherries during the brining-curing period demonstrates that the inclusion of a small amount (10%) of such contaminated fruit can cause the degradation of a large amount of cherries stored in barrels or tanks.

The source of the "cats claw" damage of cherries was not determined during 1958. The fact that many microorganisms are present at the site of this type of spoilage whereas sound fruit normally has only a small microbial population, would suggest the probability that either yeast or mold growth, with concomitant secretion of a polygalacturonase, leads to this type of contamination. How such organisms are able to penetrate the surface of the fruit originally is not known, but it is possible that the surface of some of the fruit is first damaged in some way during maturation, thus allowing microbial growth to start at this exposed site.

Of the microorganisms isolated from "cats claw" infected fresh cherries, one, or a group, of the yeasts is the most likely source of polygalacturonase production, since no evidence of mold growth was noted; and, at the advanced stage of contamination in the samples tested, a heavy mold growth would be readily visible. All mold isolated produced a heavy visible growth on the polygalacturonic acid test substrate. Phaff and Demain *(20)* have demonstrated the production of a single polygalacturonase by the yeast *Saccharomyces fragilis* which caused rapid hydrolysis of the pectins tested.

Excessive polygalacturonase activity was not found in firm fresh green or ripe cherries, or in cherries infected with cherry powdery mildew or brown rot. It is unlikely, therefore, that any appreciable polygalacturonase is formed during the maturation of cherries, or when cherries are attacked by either of these two infestations.

The inhibition of the polygalacturonase of Pectinol M solutions at pH 4.0 was found to be directly proportional to the concentration of the Nacconal NR inhibitor used at the two enzyme concentrations tested. The use of this detergent as an inhibitor of the polygalacturonase of "cats claw" infected cherries was not attempted since the polygalacturonase activity of these samples had stopped before the effectiveness of Nacconol NR as an inhibitor had been determined. When this detergent was added to firm brined Royal Anne and Bing cherries, no difference could be detected between these samples and control samples either while the cherries were in the brine, or after they were processed into maraschino cherries. The concentration of Nacconol NR used in these tests was as high as 0.5%. On the basis of the inactivation studies, a concentration of 0.025% of the detergent should cause complete inhibition of the polygalacturonase of "cats claw" infected cherries.

Proof of the safety of Nacconol NR as an additive for the inhibition of the polygalacturonase in brined cherries should be established before its general use for this purpose can be recommended. Further tests must be conducted, also, to insure that there is no deleterious effect to the cherries caused by the addition of this agent early in the brining process.

SUMMARY AND CONCLUSIONS

The "cats claw" disease of Bing and Lambert cherries was found to contain a large concentration of a polygalacturonase, and these cherries caused rapid deterioration of brined cherries when 10% by count of the infected cherries were brined with firm Bing cherries. The cherries had deteriorated noticeably within 10 days after brining. Since the pectic materials are generally conceded to be responsible for the firm texture of brined cherries, it would appear that a polygalacturonase, rather than other types of polysaccharidases, is primarily responsible for the observed softening.

The type of softening observed in these cherries was similar to that described by McCready and McComb in 1954, and to that found in commercially packed cherries from the 1957 season. This indicates that the type of spoilage which resulted in the softening of brined cherries during 1957 was probably identical to that caused by "cats claw" contaminated cherries.

Little further deterioration was observed in these brined cherry samples after the enzyme activity had stopped. The period of breakdown was found to parallel the time of polygalacturonase activity.

No polygalacturonase activity could be demonstrated in firm ripe cherries, nor in cherries infected with cherry powdery mildew or brown rot. None of these appears to be involved in the softening of cherries during the brining and storage period.

The alkyl aryl sulfonate detergent Nacconol NR was found to be an effective inhibitor for the polygalacturonase in Pectinol M in cherry brine at pH 2.6 and in buffer solutions at pH 3.0, 3.6 and 4.0. Inhibition of this enzyme by Nacconol NR at pH 4.0 was found to be directly proportional to the concentration of the detergent added for 0.1 and 0.2% Pectinol M solutions. This type of inhibition is typical of noncompetitive enzyme inhibitors.

The detergent Nacconol NR did not cause any deleterious effect when added to firm brined Bing or Royal Anne cherries at a concentration of 0.5% two weeks before processing into maraschino cherries. The effect caused by adding Nacconol NR to cherries at the time of brining was not determined and this should be done before it is used as a general inhibitor of polygalacturonase.

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Factors Affecting Decay of Prepackaged Spinach[a]

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HARVESTED LEAFY VEGETABLES such as spinach are extremely susceptible to attack by bacterial soft rot organisms. After carefully investigating the spinach soft rot problem, Smith *(10)* reported that *Erwinia caratovora* (Jones) Holland is the principal organism involved. However, other species of bacteria, as well as several species of fungi and yeast, have also been shown to produce pectinolytic enzymes capable of decomposing plant tissue *(13)*.

Smith *(10)* obtained experimental evidence that infection may take place in either injured or uninjured leaves, but that wounding facilitates more rapid decay. He further noted that high temperature and humidity favor the development of decay.

Several workers *(4, 5, 9, 14)* have pointed out that prepackaged spinach is particularly subject to rapid decay, due to injury to leaves during the prepackaging operation and bacterial inoculation of noninfected leaves during the washing process. Occurrence of soft rot limits the feasibility of commercial prepackaging, especially during the warm summer months.

To overcome the harmful effects of high temperature, refrigeration has been widely used to reduce the incidence of soft rot in prepackaged spinach. In commercial practice, excess wash water is commonly removed by centrifuging the spinach prior to bagging. However, despite refrigeration, centrifugation, and careful handling, the shelf life of prepackaged spinach is often limited to a very few days.

To prolong the period of salability, several workers have investigated the possibility of adding bacteriostatic chemicals to the wash water. Lieberman *et al (8)*, and later Friedman *(6)*, tried chlorine. Although bacterial populations were reduced, shelf life was not markedly increased. Adding antibiotics to the wash water was first tried by Smith *(11, 12)*. Other workers, including Brody and Francis *(3)* and Koch and Carrol *(7)*, have tried similar experiments, using several antibiotics at different concentrations. Their results indicate that the antibiotics streptomycin, oxytetracycline, and chlortetracycline are each capable of inhibiting soft rot development in prepackaged spinach.

In 1958, Becker, Goodman, and Goldberg *(2)* reported that the antibiotics oxytetracycline, chlortetracycline and streptomycin sulfate, used at low concentrations, showed promise as supplements to refrigeration by improving the appearance and prolonging the shelf life of prepackaged spinach. Results of these experiments indicated that all three antibiotics reduced the amount of decay and improved the appearance of the product under conditions that resulted in rapid decay. When soft rot developed slowly, the antibiotics had little or no effect in inhibiting decay development. By the use of serial dilution plate counts, it was shown that the antibiotics markedly reduced the number of microorganisms present.

Although residues were detected immediately after treatment with oxytetracycline or chlortetracycline, no antibiotic activity was detected when samples were cooked for 3 minutes in boiling water. With streptomycin, residues were detected immediately after treatment and still remained after cooking.

Low concentration antibiotic dips appear to be effective in reducing the amount of decomposition only under conditions of rapid decay. Therefore it was felt more information is needed to ascertain when antibiotics might profitably be used to inhibit soft rot development. The present report is based on work to determine what factors affect the rate of decay of prepackaged spinach.

MATERIALS AND METHODS

The spinach used in the experiments was obtained in center-iced bushel baskets through commercial wholesale channels. Undamaged, disease-free leaves were washed, drained, and submerged for 1 min in an 18-hr nutrient dextrose broth culture of *E. caratovora*, diluted 1:1000 in water. They were then drained for 20 min in screen-bottom trays. To approximate an antibiotic dip, used concurrently on other spinach in some of the experiments, the spinach was submerged in water for 1 min. Excess liquid was removed by the spin dry cycle of a standard automatic clothes washer. Samples weighing 110 g each were packed in perforated polyethylene bags and stored at 7.2° C (45° F).

Samples were taken at regular intervals to determine the amount of decay present. Each leaf was visually inspected and placed in one of 4 decay classes, based on the amount of decay. The approximate percentage of decay that each class represents is: *very slight* (2% or less); *slight* (15% or less); *medium* (50% or less); *heavy* (more than 50%). For determining the extent of decay, the classes were assigned the factors 1, 2, 3, and 4, respectively. After the samples from each treatment had been sorted according to this classification, the leaves in each class were weighed, and the weight was multiplied by the assigned factor. The adjusted figures for the 4 classes were then totaled. The resulting sum was divided by the total amount (in grams) of spinach in the treatment. This provided an arbitrary decay rating for each treatment.

In 16 of the 24 experiments, serial dilution plate counts were made at regular intervals to determine the microbial population. Samples of 100 g each were macerated for 2 min in 200 ml of sterile distilled water in a Waring blender. A

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10 ml aliquot of the resulting slurry was then serially diluted and plated, following accepted milk-testing procedures *(1)*. Nutrient dextrose agar was employed to ascertain the number of bacteria present; Littman media containing 50 mmc/ml of oxytetracycline was used to differentiate yeast and fungi.

The procedure described above was modified in experiments 22, 23, and 24 to include two additional treatments. Besides dipping spinach in a suspension of *E. caratovora* diluted 1:1000 in water, spinach was also submerged in a 1:100 dilution of the inoculum. The third treatment was obtained by simply sorting, washing, centrifuging and bagging untreated spinach leaves. Samples were periodically removed from cold storage and examined to establish decay ratings for each treatment. Plate counts were also made.

When it was noted that the total microbial populations were not necessarily proportional to the rate of decay, an effort was made to determine the percentage of soft rot organisms present. Since *E. caratovora* and other decay-producing organisms decompose potato as well as spinach, it was decided to use potato slices as a selective test. Surface-sterilized Irish potatoes were aseptically sliced and placed in petri dishes on moist, sterile filter paper. A single bacterial colony was then picked from or beneath the surface of the nutrient dextrose agar with a culture needle and stabbed into the center of a potato slice. Thirty to 50 individual colonies were selected at random after each plating for each of the three treatments in experiments 22, 23, and 24. The same procedure was followed in the cases of yeast and fungi. The petri dishes were examined at 24 and 48 hours to ascertain which colonies incited decay.

Work was carried on to determine the per cent moisture content of fresh, untreated spinach. Samples were dried to constant weight at 65° C.

Data were also secured on the amounts of nitrogen, phosphorus, potassium, calcium and magnesium in fresh spinach. The percentage of each element was obtained by spectrographic analysis of air-dried samples.

RESULTS

The number of days necessary for artificially inoculated (dilution 1:1000) prepackaged spinach to reach decay ratings of 100, 150 and 200 are presented in Figure 1. These data on the rate of soft rot development reveal that samples decayed more rapidly during the summer months than during the rest of the year.

The population of microorganisms in 100-g samples of spinach was taken 1, 4, 8 and 12 days after treatment (Figure 2). In every experiment the results show that the number of yeast and fungi combined is relatively low in comparison to number of bacteria. There is some evidence of a direct relationship between number of bacteria and rate of decay, Figure 1, or an inverse relationship between population and number of days necessary to reach decay rating of 100, 150, and 200. This relationship is probably most evident in the case of the bacterial populations in experiments 14 through 24, sampled at 1 and 4 days after treatment. With the yeast and fungi, a relationship between number of organisms and rate of decay does not appear to be clear cut or constant.

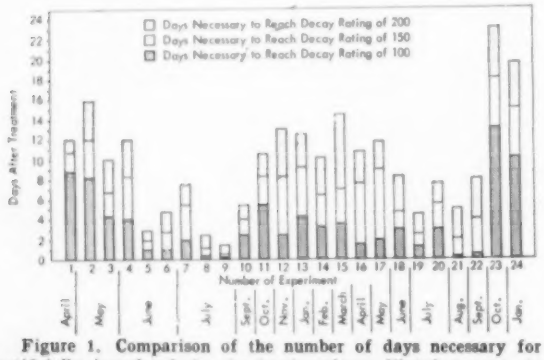

Figure 1. Comparison of the number of days necessary for artificially inoculated (18 hr broth culture diluted 1:1000 in water) prepackaged spinach to reach decay ratings of 100, 150, and 200 when stored at 45° F.

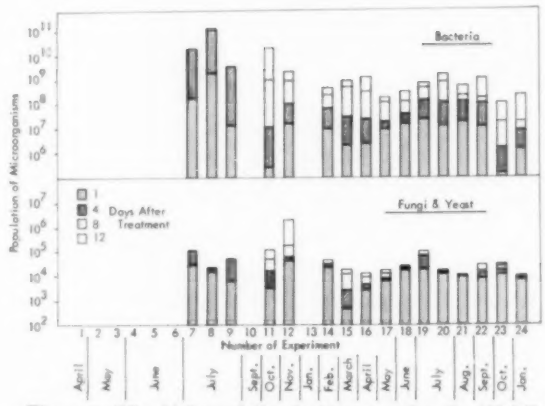

Figure 2. Microbial population in 100-g samples of artificially inoculated spinach 1, 4, 8 and 12 days after treatment.

When samples of artificially inoculated (1:100) and uninoculated prepackaged spinach were examined at various intervals, decay ratings revealed that the spinach dipped in the bacterial suspension decayed more rapidly than did the uninoculated spinach. Figure 3 illustrates the results obtained in experiments 22, 23 and 24.

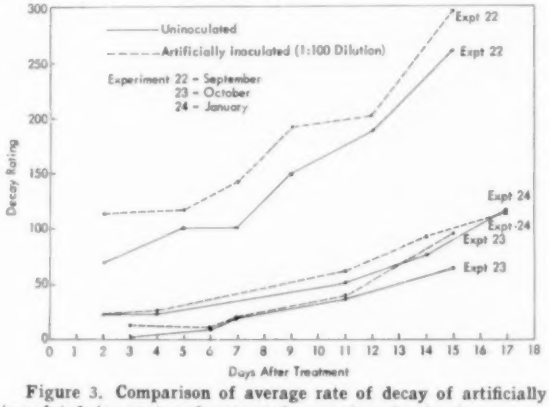

Figure 3. Comparison of average rate of decay of artificially inoculated (1:100) and untreated prepackaged spinach.

The populations of yeast and fungi obtained from 100-g samples of uninoculated spinach and the number of bacteria obtained from artificially inoculated (1:100) and uninoculated spinach are shown in Figure 4. In each experiment, the bacterial population increased steadily until the termination of the experiment, while the number of yeast and fungi remained relatively constant.

Table 1 gives the percentages of microorganisms capable of inciting decay of potato tuber slices. Although the percentage of decay organisms seems to vary from one sampling to the next, the curves obtained by plotting the calculated actual number of such organisms (Figure 5) closely resemble those of the total microbial population (Figure 4).

Table 2 contains data on the amounts of nitrogen, phosphorus, potassium, calcium, and magnesium in air-dried samples. There apparently was little difference in the amount of moisture from one lot of spinach to the next, but the content of the 5 elements varied considerably.

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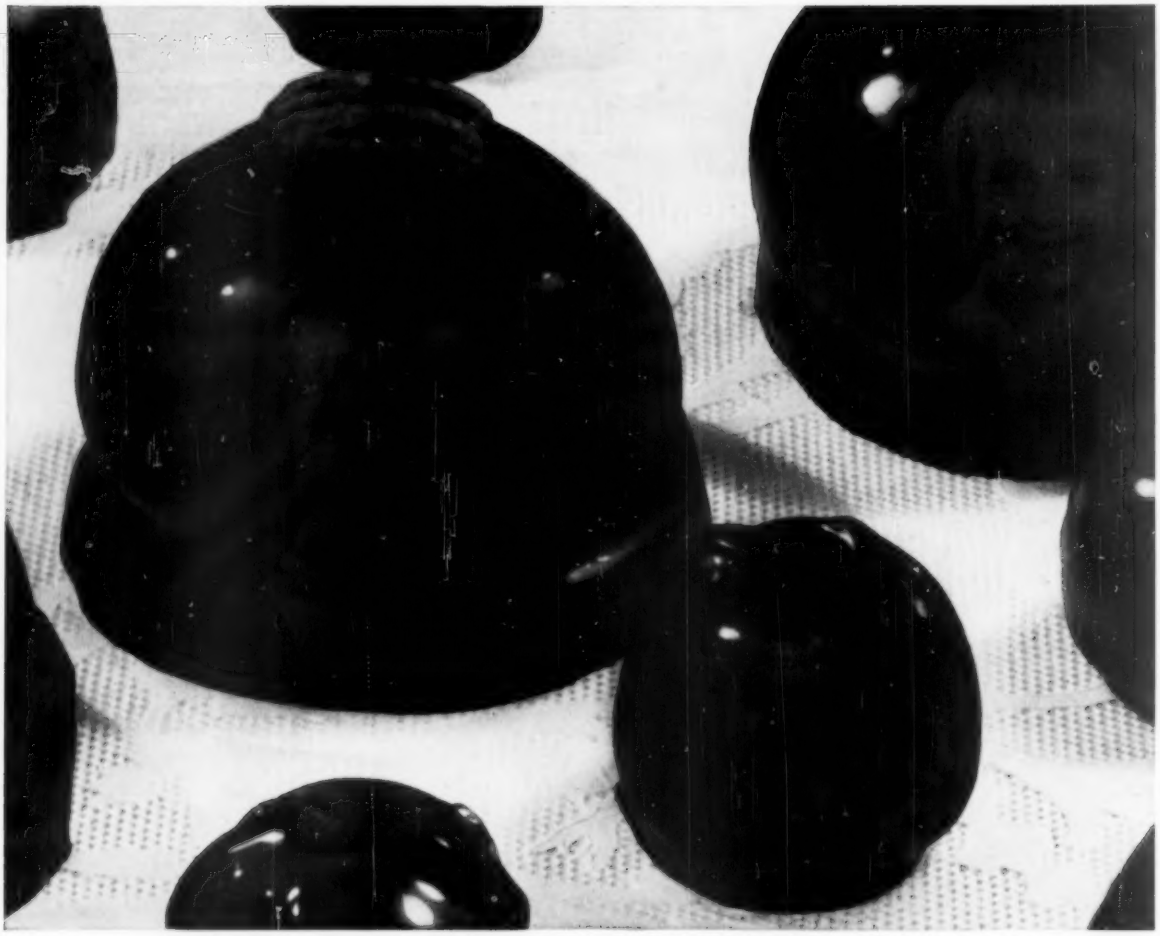

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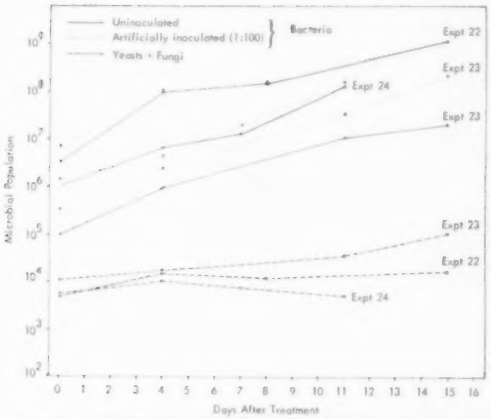

Figure 4. Microbial population contained in 100-g samples of artificially inoculated (1: 100) and uninoculated prepackaged spinach.

DISCUSSION

During this study, it was noted that prepackaged spinach decayed most rapidly during the summer months (Figure 1). Since the spinach was handled the same and stored at 7.2° C (45° F) in all experiments, the results suggest either that the population of soft rot organisms was greatest during the summer, or that the spinach was predisposed to decay by warm temperature, rapid growing conditions, or some other factor.

TABLE 1

Per cent of total bacteria demonstrated to produce soft rot of potato tuber slices

Experiment number	Days after treatment	Per cent of total found to induce decay		
		Bacteria		Fungi and yeasts
		Uninoculated	Artificially inoculated	
22	0	15.6	20.0	25.9
	4	9.1	18.4	17.8
	8	18.5	26.7	37.0
	15	19.7	18.2	85.0
23	0	14.3	86.7	44.1
	4	23.1	44.4	41.7
	11	27.3	59.8	35.4
	15	15.8	25.8	34.6
24	0	27.0	20.0	63.3
	4	25.0	22.6	43.9
	7	28.6	26.5	30.0
	11	31.2	41.9	40.0

When the data in Figure 1 are compared with the population of microorganisms, Figure 2, some evidence of a direct relationship between the number of bacteria and rate of decay can be seen.

It was also observed that artificially inoculated spinach decayed more rapidly than uninoculated (Figure 3). If the data (Figure 3) are compared to the microbial population (Figure 4), it is evident that rate of decay is proportional to number of bacteria. However, in the case of yeast and fungi, such a relationship is not evident.

TABLE 2

Relative amounts of nitrogen, phosphorus, potassium, calcium and magnesium in air-dried samples of spinach, and the average per cent of moisture in fresh spinach

Experiment	Per cent moisture[2]	Per cent of dry matter[1]				
		Nitrogen	Phosphorus	Potassium	Calcium	Magnesium
22	13.02	6.03	0.86	7.46	0.72	0.67
23	12.78	4.88	0.54	9.24	0.64	0.93
24	12.58	5.14	.....	6.92	2.04	0.71

[1] Information obtained by spectrographic analysis of air-dried samples.

[2] Leaves and petioles dried to constant weight at 65° C.

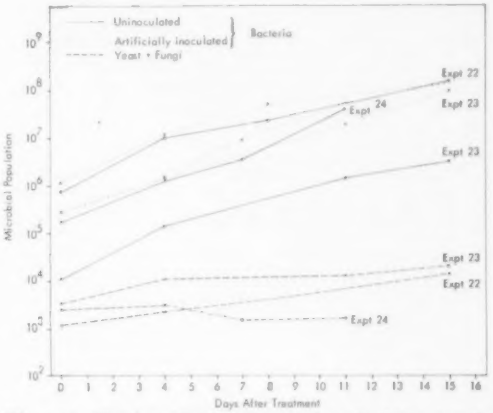

Figure 5. Number of soft rot organisms in 100-g samples of artificially inoculated (1: 100) and uninoculated prepackaged spinach.

Since a great number of the microorganisms obtained from spinach undoubtedly do not incite decay, there is some question as to the value of data based on the total microbial population. To determine what percentage of the total number of microorganisms was capable of causing decay, potato tuber slices were used as a selective test. It is probably safe to assume that most of the organisms which were able to decompose potato tissue could cause decay of spinach, pectinolytic enzymes being involved in both cases. When the calculated number of these decay-producing organisms (Figure 5) and the total microbial population (Figure 4) are plotted graphically, a marked similarity is seen between the two sets of figures.

The data which have been presented thus indicate that total bacterial population and the population of actual soft rot bacteria are directly proportional to rate of decay. However, this does not appear to be the complete explanation. In experiment 23, there were 10 times more soft rot bacteria in the artificially inoculated spinach than in the uninoculated; yet there was little difference in rate of decay between the 2 treatments. On the other hand, in experiment 22, the soft rot bacterial population varied little between treatments, whereas there was marked difference in rate of decay. This suggests that other factors besides the actual number of decay-producing bacteria influence soft rot development.

Although yeast and fungi must play some role in controlling rate of decay, results of the present study show no clear relationship between number of organisms and decay development. It may be significant that the yeast and fungi were few in number compared to the number of bacteria, and their effect on rate of decay might have been masked. It is possible that under conditions of low bacterial populations, their effect would be more pronounced.

As Table 2 shows, the content of the 5 elements determined in this study varied considerably. The nitrogen content is particularly interesting, in that it is a common observation among commercial producers that spinach grown on land high in nitrogen decomposes readily. In experiments 22, 23, and 24, there is a relationship between rate of decay and amount of nitrogen. Although these figures are too few to constitute proof, they suggest that the nitrogen content of spinach tissue does have an effect on soft rot development.

SUMMARY AND CONCLUSIONS

Previous work showed that, under conditions of rapid decay, 25 meg/ml solutions of oxytetracycline, chlortetracycline and streptomycin sulfate retarded the development of soft rot and reduced the population of bacteria in prepackaged spinach. Data from the present study indicate that spinach grown during the hot summer months is more susceptible to decay than that grown at other times of the year.

Experimental evidence has shown that total bacterial population and the population of actual soft rot bacteria are directly proportional to rate of decay. There is no such relationship in the case of yeast and fungi. Thus, rate of decay appears to depend on the bacterial population of the spinach. However, other factors, such as the nitrogen content of the leaves, may also play a part in determining the decay rate, either by affecting the microorganisms or by predisposing the spinach to attack.

Evidence obtained from this study suggests that antibiotics used at low concentrations have practical applications in the preservation of leafy vegetables such as spinach. It is felt that these chemicals would be particularly effective in reducing the number of bacteria during warm weather, when soft rot development is most rapid.

To date, the Federal Drug Administration has not approved the use of antibiotics in the preservation of harvested vegetables. However, since spinach is usually eaten after it is cooked, heat labile antibiotics such as oxytetracycline and chlortetracycline may in the future find a place in the preservation of harvested leafy vegetables.

Acknowledgment

This study was supported by funds from United States Public Health Service Grant-in-aid RG 4407, National Institutes of Health, Bethesda, Maryland.

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The Collapsible Polyethylene Tube as a Food Package[a]

J. D. Kemp, R. M. Ballantyne, A. J. Ducker, and J. W. Haynes

Defence Research Medical Laboratories, Toronto, Ontario, Canada

(Manuscript received September 25, 1959)

MANY TYPES of military field rations contain spreads such as jam, jelly and peanut butter. Individual rations require a light weight package containing about two ounces of such a spread. Standard commercial packages are usually cans or glass jars which contain quantities sufficient for several meals. The smallest sanitary can commercially available in Canada holds four ounces of jam and the only individual portion package—the small, formed plastic cup used by restaurants and airlines—is too fragile for military use.

To satisfy the need for a suitable package—one that is light in weight, will protect the food satisfactorily for two years at 70° F, will withstand a moderate degree of rough usage, is capable of being reclosed, and will not present a serious damage hazard to other packages with which it might be assembled—the use of collapsible polyethylene tubes was considered.

The tubes proved to have considerable strength. A filled and properly sealed tube laid on its side would support one hundred and fifty pounds weight without rupturing; therefore the tubes were not regarded as being a serious breakage risk in field rations.

A series of storage experiments were conducted in the cold (0° F), at a normal room temperature (70° F) and at an elevated temperature (100° F). The results of these experiments are described below.

EXPERIMENTAL

Materials

Polyethylene tubes. The tubes used were of nominal 2- and 2.4-fluid oz capacity, fabricated from low density polyethylene. The walls were extruded tubing with a thickness of 0.010–0.012 in and an outside diameter of approximately $1\frac{3}{16}$ in. The shoulder and nozzle were moulded onto the extruded wall, the shoulder thickness being approximately $\frac{3}{64}$ in. The wall length was $4\frac{1}{2}$ in for 2-oz tubes and $5\frac{1}{8}$ in for 2.4-oz tubes. The latter were coated internally with a polyvinylidene chloride type resin. This coating, estimated at 0.0003 in in thickness, stopped approximately $\frac{1}{4}$ in from the open end of the tube to avoid fouling of the heat-seal closure area. The tube nozzles had an inside diameter of $\frac{3}{16}$ in and were fitted with moulded screw caps. The tubes weighed approximately 0.2 oz each, including cap.

Foods. Five foods were used: pure grape jelly, pure raspberry jam, white No. 1 pasteurized honey, homogenized peanut butter, and creamery butter.

Procedure

Phase I. Sixty unlined 2-oz tubes were filled with each of the following foods: grape jelly, raspberry jam, honey (each in the amount of 2.6 oz per tube), and peanut butter (2.3 oz per tube). The tubes were sealed, weighed accurately, and placed in storage under four conditions, viz., 0° F, 70° F, 100° F, and a temperature cycle which alternated monthly between 0° F and 70° F. All tubes were weighed periodically; sample tubes were withdrawn at regular intervals for 1 year from those stored at 100° F, for 2 years from those stored at 0° and 70° F, and for 18 months from those undergoing cycling. The tubes were examined visually and the foods assessed for colour, flavour and texture using the samples of food kept at 0° F as controls. Peroxide value analyses *(2)* were performed on peanut butter to detect oxidative changes.

Phase II. It was considered desirable to obtain direct comparisons among the foods stored in tubes and stored in standard types of packaging. Forty unlined 2-oz tubes and forty lined 2.4-oz tubes were filled with each of the following foods: grape jelly, raspberry jam, and honey (each in the amount of 2.6 oz in unlined tubes, 3.0 oz in lined tubes). The tubes were sealed, weighed accurately, and placed in storage at 100° F. Forty 3-oz brown glass bottles with lined screw caps, and forty 211 x 105 internally-enamelled tinplate cans were also filled with each food and stored at 100° F. All tubes were weighed monthly. Sample tubes, bottles and cans were removed monthly during the 6 months of the test, the tubes were examined and the colour, flavour and texture of the foods compared. Quantities of the foods in their original containers were held at 35° F and used as controls.

Phase III. During *Phase I*, the peanut oil was found to penetrate the unlined tubes in a few months. To compare the resistance of the 2 types of tubes to permeation by oils, 3 unlined 2-oz tubes and 6 lined 2.4-oz tubes were filled with each of the following foods: peanut butter and creamery butter. The tubes were sealed and placed in storage at 100° F. Samples were removed monthly during the 6 months of the test for evaluation of food and package.

Because of the relatively small number of tubes used for each food in these experiments, machine filling was not practical and all tubes were filled by hand. The honey and butter were liquefied for filling; the jam, jelly and peanut butter were dispensed through an icing tube normally used for cake decoration.

Tubes in *Phase I* were sealed in a portable, hand-operated machine; those in *Phase II* were sealed in a production machine operating at factory speed; those in *Phase III* were sealed in a jaw-type heat-sealer using a sheet of polyester film to prevent sticking of the polyethylene to the heated jaws. All tubes, bottles and cans of food were stored without being heat-processed.

The constant temperature storage rooms were controlled for temperature only. Forced air circulation permittted control to within ±3° F.

RESULTS AND DISCUSSION

Physical changes. The tubes of jam, jelly and honey lost weight as shown in Figures 1 to 3; unlined tubes of peanut butter gained weight as shown in Figure 4.

Since in the storage areas only temperature was controlled, relative humidity varied seasonally—30 to 60% relative humidity in summer, 15 to 60% *r* in spring and fall, and 5 to 40% *r* in winter at a room temperature of 70° F. These seasonal changes are shown most clearly by the 70° F curve of Figure 1; the flat portions represent low rates of weight loss

[a] Issued as Defence Research Medical Laboratories Report No. 174–7, PCC Project No. D50-67-20-03.

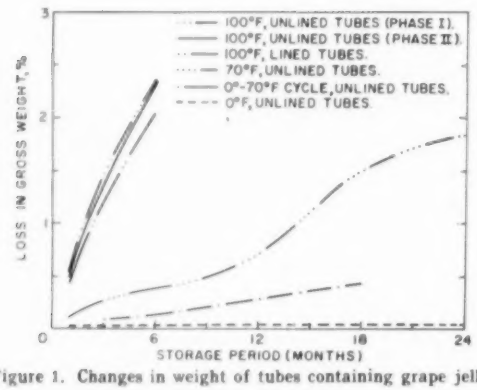


Figure 1. Changes in weight of tubes containing grape jelly.

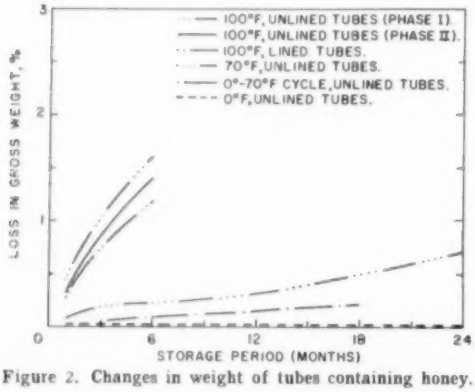


Figure 2. Changes in weight of tubes containing honey.

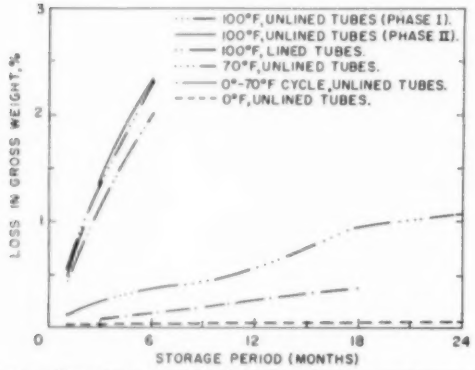


Figure 3. Changes in weight of tubes containing raspberry jam.

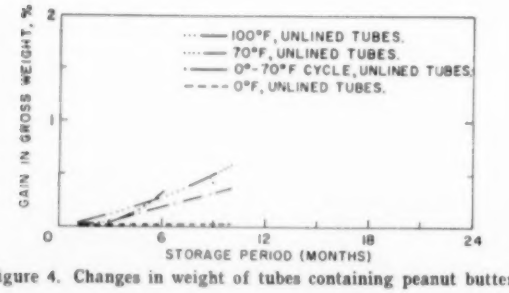


Figure 4. Changes in weight of tubes containing peanut butter.

during the spring and summer months when indoor relative humidity was highest.

At 100° F, weight losses from unlined tubes were approximately 7 times as great as at 70° F for the same storage period, and there was very good agreement between the results of *Phase I* and *Phase II*. Lined tubes were slightly more effective than unlined tubes in controlling weight loss.

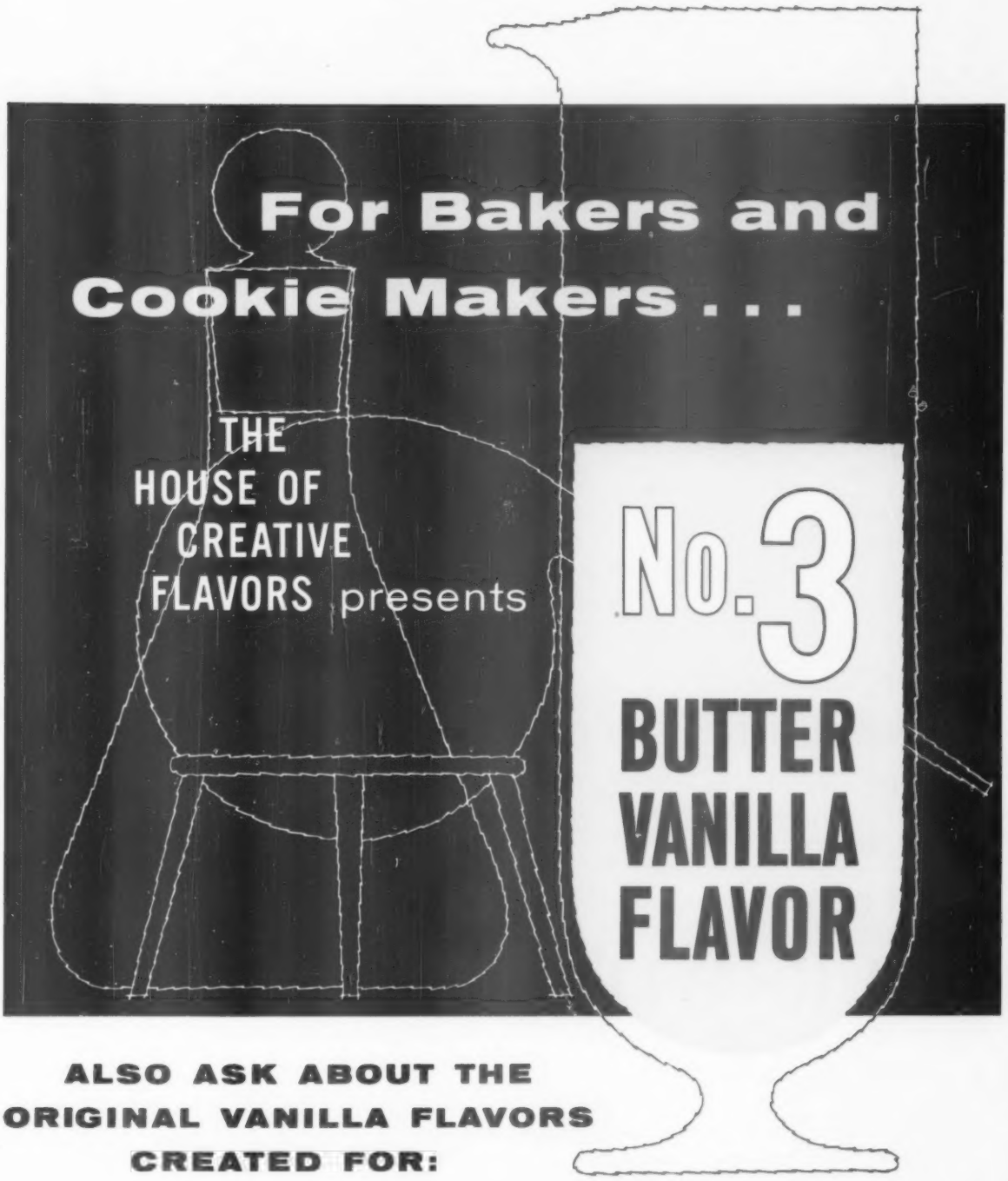
There were no apparent physical changes in the tubes themselves at any of the 4 storage temperatures, and there was no evidence that the tubes had leaked. It was suspected that weight losses were due to the permeation of moisture through the polyethylene, particularly since seasonal changes in ambient relative humidity had a considerable effect on weight losses at 70° F. Results of an empirical test with similar tubes filled with water supported this view.

Unlined tubes of peanut butter in *Phase I* showed increasing gains in weight. Tube surfaces became oily within 3 months at 100° F and within 6 months at 70° F. The peanut butter developed a rancid flavour and showed a substantial increase in peroxide value with time (see Table 1). Since polyethylene is permeable to oxygen *(1)* it is concluded that the weight gains were the result of oxygen transfer from the atmosphere to the product. Tubes of peanut butter were discarded after 10 months' storage because of excessive oiliness of the tube surfaces. In *Phase III*, unlined tubes containing peanut butter and creamery butter stored at 100° F developed surface oiliness within 2 months, supporting earlier observations. In contrast, lined tubes did not develop surface oiliness even after 6 months under the same storage condition. There was a tendeney for the lining to loosen around the neck of the tube but adhesion to the tube walls remained satisfactory.

Food flavour and appearance. Jam and jelly stored at 70° F for one year in unlined tubes developed a brown colour and a slightly caramel flavour which continued to increase until the products were of only borderline acceptability when sampled after 2 years. Honey was light amber after 1 year and amber after 2 years at 70° F, but the flavour and aroma were still good. About one-third of the honey in each tube had crystallized. When peanut butter was removed from the storage programme at 10 months, a slightly rancid flavour and slight darkening of the colour had developed.

As would be expected, all foods stored at 0° F retained their normal colours and flavours throughout 2 years. Honey showed a tendency to develop a coarse crystalline texture; the textures of the other foods were unchanged.

Jam, jelly and honey stored under cyclic conditions deteriorated more slowly than those kept continuously at 70° F; their flavour and appearance after 18 months of cyclic storage were comparable to those after a year at 70° F. There was no difference in either the colour or the flavour of peanut butter samples stored for 10 months at 70° F and under cyclic conditions.



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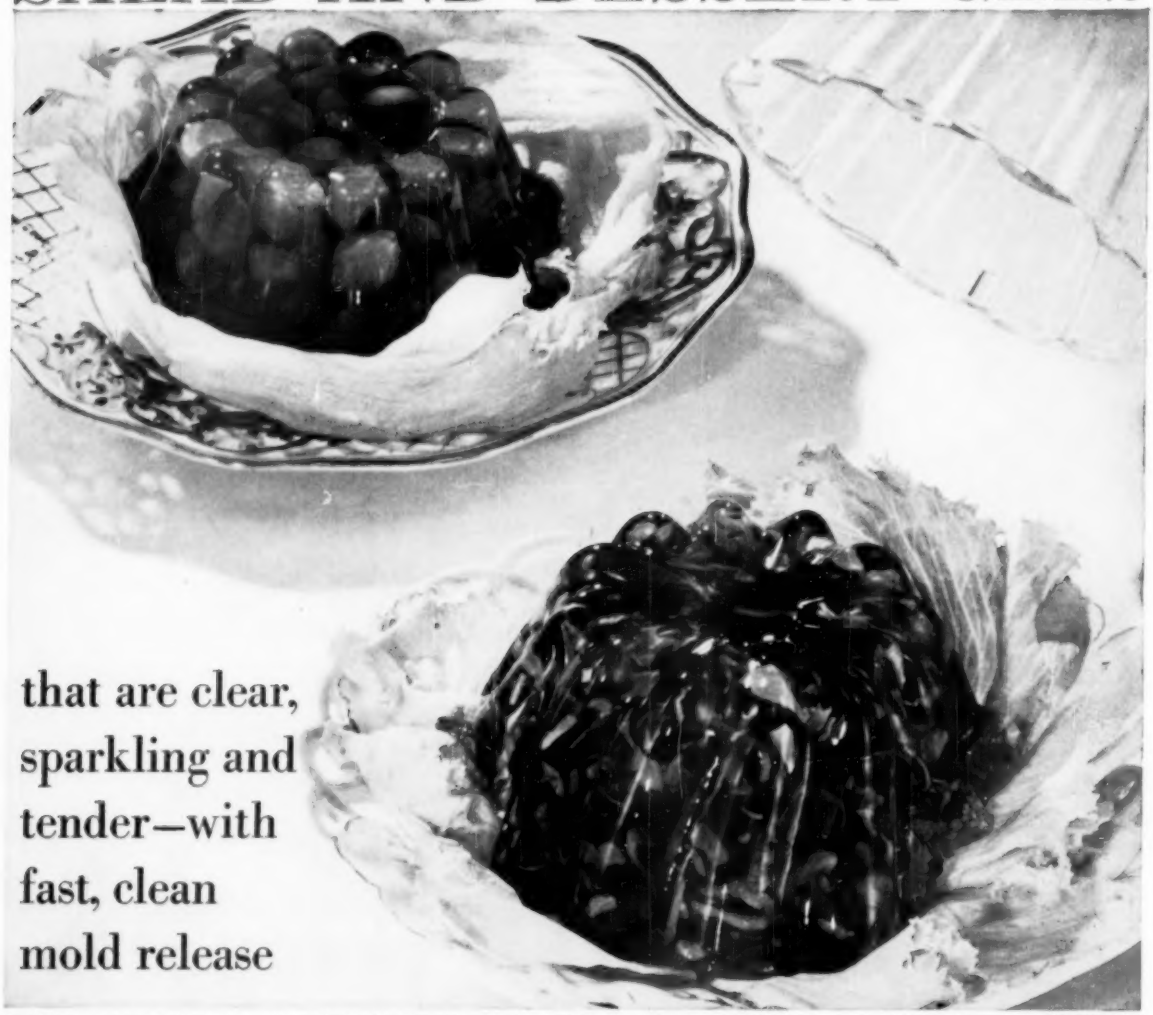

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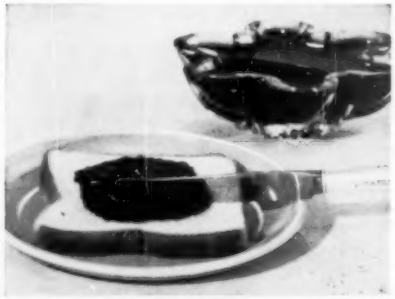

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132B

Storage for 6 months at 100° F is regarded as a severe test for foods and is employed when a general appraisal of storage characteristics must be obtained in a short time or when storage life in a tropical environment must be predicted. After 6 months at 100° F, the jam and jelly in unlined tubes were brown in colour, had developed a caramel flavour and were judged to have only borderline acceptability. The honey had partially crystallized and had developed a light brown colour and a caramel flavour; it was no longer of good quality but it was not unpalatable as a spread. The peanut butter had become darker in colour, the flavour moderately rancid and the oil had separated from the solids; the peroxide value analyses of Table 1 indicate the extent to which oxidative rancidity had progressed.

It was apparent that the unlined tubes were not suitable for packaging peanut butter though they were quite satisfactory for honey and held promise of being suitable for jam and jelly. Deteriorative changes occurred and were generally comparable at 6 months at 100° F and 2 years at 70° F.

Deterioration of the type observed—browning and caramelization—is known to occur spontaneously in fruit products *(3)*, so that comparisons with other packages, viz., glass bottles and metal cans, were undertaken. Food quality was compared monthly for 4 packages; results are summarized in Tables 2 to 4. None of the foods showed significant deterioration in comparison with the controls (held at 35° F) at the one month examination. Assessments were not made at 6 months because the foods in all packages had deteriorated to such an extent that comparisons were of little value.

TABLE 1

Peroxide value analyses for peanut butter

Storage temperature, 0° F	Length of storage, months	Peroxide value[1]
0°	3	1
	6	2
	10	8
0°–70° cycle	3	2
	6	62
	10	59
70°	3	5
	6	56
	10	58
100°	3	6
	6	135

[1] Rancidity was detectable organoleptically at a PV of 35.

TABLE 2

Comparative assessment of raspberry jam in various packages

Package	Duration of storage at 100° F			
	2 months	3 months	4 months	5 months
Glass bottles and metal cans	Slight browning compared to the control; fresh raspberry flavour lost; caramelization detected.	Red-brown colour; little raspberry flavour; distinctly caramel.	Brown colour; sweet, distinctly caramel; raspberry flavour not recognized.	Decidedly brown colour; sweet, distinctly caramel, no raspberry flavour.
Lined tubes	Appearance and flavour similar to jam in bottles and cans.	Brown colour with slight redness; flavour similar to jam in bottles and cans.	Colour and flavour generally similar to jam in bottles and cans; slight off-flavour in jam at the top of the tubes.	Colour similar to jam in bottles and cans; sweet, caramel flavour with a slight astringent off-flavour.
Unlined tubes	Colour similar to jam in cans and bottles; quite definite caramel flavour.	Brown colour; caramel flavour; distinct "burnt paraffin" off-flavour in jam at top of tubes but barely noticed in well-mixed sample of entire contents.	Colour slightly more brown than in other packages; sweet caramel flavour with a "burnt paraffin" off-flavour throughout.	Colour similar to that in other packages; decided caramel flavour with a "bitter, burnt paraffin" off-flavour.

TABLE 3

Comparative assessment of grape jelly in various packages

Package	Duration of storage at 100° F			
	2 months	3 months	4 months	5 months
Glass bottles and metal cans	Dull red; very slight caramelization observed.	Dull red; faint grape flavour, mainly sweet and caramel.	Brown; faint grape flavour, pronounced caramelization.	Brown; sweet, caramel flavour.
Lined tubes	Red-brown; very slight caramelization observed.	Colour and flavour similar to jelly in bottles and cans.	Slightly lighter brown than jelly in bottles and cans; sweet, caramel flavour.	Slightly lighter brown colour than, but flavour similar to jelly in bottles and cans.
Unlined tubes	Slightly more brown than jelly in lined tubes; slight off-flavour observed.	Dull red-brown; caramel flavour; distinct "burnt paraffin" off-flavor in jam at top of tubes but barely noticed in well-mixed sample of entire contents.	Colour similar to jelly in lined tubes; "burnt paraffin" off-flavour in addition to caramelization.	Colour similar to jelly in lined tubes; "burnt paraffin" off-flavour in addition to caramelization.

TABLE 4

Comparative assessment of honey in various packages

Package	Duration of storage at 100° F			
	2 months	3 months	4 months	5 months
Glass bottles and metal cans	Slightly amber flavour similar to the control.	Light amber; flavour similar to the control.	Amber; flavour resembled a stronger amber honey.	Amber; flavour had become still stronger.
Lined tubes	Slightly amber; some crystallization; flavour similar to the control.	Slightly deeper amber than honey in bottles or cans; flavour similar to the control.	Slightly deeper amber than, but flavour similar to honey in bottles or cans; cloudy.	Deep amber; cloudy due to crystallization; sharp caramel flavour predominated.
Unlined tubes	Similar in flavour and appearance to honey in lined tubes.	Similar in flavour and appearance to honey in lined tubes.	Slightly deeper amber colour than, but flavour similar to honey in lined tubes; cloudy.	Similar in flavour and appearance to honey in lined tubes.

Jam and jelly in the unlined tubes developed an off-flavour in 3 months; such flavours were not observed in *Phase I* which suggests the possibility of difference in fabrication between the 2 sample lots of tubes. In lined tubes, off-flavours were observed only in the raspberry jam, and not to a serious degree until 5 months. There was little difference among the glass bottles, metal cans or lined tubes of either jam, jelly or honey during the first 3 months of storage; thereafter, foods in the bottles and cans were slightly superior.

The peanut butter and creamery butter stored at 100° F in lined tubes maintained a satisfactory flavour for 3 months but by the end of 6 months both foods were rancid.

SUMMARY

Raspberry jam, grape jelly, honey, peanut butter and creamery butter were used in a series of storage studies to evaluate lined (i.e., coated with polyvinylidene chloride resin) and unlined collapsible polyethylene tubes as food containers.

Lined tubes were found to be more suitable than unlined tubes because of a lesser tendency to produce off-flavours and because of their impermeability to oils. This impermeability indicated their potential suitability as packages for oily foods such as butter and peanut butter, but the oxygen transmission characteristics of the tubes should be investigated more fully.

During comparative tests at 100° F the storage life of jam, jelly and honey in lined tubes was generally comparable to that in bottles or cans for periods up to three months; thereafter, the quality of foods in bottles and cans was slightly superior.

Acknowledgment

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Doneness of Frozen, Defrosted Turkey Halves Roasted to Several End Point Temperatures [a, b, c]

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ONE OF THE MAJOR PROBLEMS encountered in roasting turkey has been lack of a satisfactory guide for determining the end point of roasting. Workers have depended on such indications of doneness as the feel of the flesh when touched with the fingers, tenderness of the meat when speared with a fork, or ease of manipulating a joint *(1)*. Birds also have been cooked for a specific number of minutes per pound, to certain end point temperatures, or by a combination of time and internal temperature *(2, 3, 4)*.

Before a reliable and uniform research method to roast turkey can be formulated, various procedures for determining end points need further examination. The purpose of this study was to investigate certain end point temperatures in both breast and thigh to determine which would produce most consistently an optimumly done bird.

EXPERIMENTAL

The first year this study was conducted the turkey halves were roasted to internal temperatures of 85° and 90° C in the breast *(pectoralis major)* and thigh muscles. End point temperatures of 85° and 90° C in the breast and 90° and 95° C in the thigh were used the second year.

Each year, 20 frozen Broad Breasted Bronze turkey hens of similar age and history were purchased as a lot from a processing plant and held in frozen storage at 0° F. Frozen turkeys were halved lengthwise and wrapped in aluminum foil (0.0015 gauge). Weights of the halves ranged between 7 and 8 lb the first year and 5½ and 6½ lb the second year.

Birds were cooked according to a split-plot design the first year and a randomized complete block design the second; in each case, a block consisted of 4 halves from 2 turkeys. Thigh temperatures were used as the end points of cooking in the 2 halves of one bird, and breast temperatures in the 2 halves of a second bird the first year; whereas, the second year end point temmperatures of 85° and 90° C in the breast and 90° and 95° C in the thigh were assigned at random to the 4 halves of each group of 2 turkeys.

Thermometers were inserted midway between the medial and lateral side at the middle of the *pectoralis major* and thigh muscles. Thermometers in the *pectoralis major* were at a 90° angle and those in the thigh at a 50° angle. Thermometer bulbs were midway between the dorsal and ventral sides of the muscles.

Turkey halves were defrosted at room temperature for 18 to 21 hr prior to being roasted at 163° C (325° F). Internal temperatures of the *pectoralis major* and the thigh muscles were recorded every 20 min after reading thermometers through an oven door window. Volatile, dripping and total cooking losses were determined.

An experienced panel judged the second year turkeys for degree of doneness before carving and soon after the birds were removed from the oven. Doneness scores for second year turkeys were based on a 7-point scale with the optimum represented by the mid-point, 4. The top of the scale, 7, described the sample as very overdone; the bottom of the scale, 1, rated the sample as very underdone. A 5-point scale with a mid-point of 3 was used the first year.

The birds were cooled approximately 2 hours, and the *gluteus primus* and *semitendinosus* muscles of the thigh and the *pectoralis major* muscle were removed for organoleptic tests. Judges received samples from the same position of the muscles at each judging period. The juice that exuded from the birds during carving was collected and poured into coded bottles for scoring. In the first year, one palatability committee scored both light and dark meat; the second year, one committee scored the light meat and another, the dark meat. Both groups scored the juice for degree of doneness, and used a range of 7 points to score flavor, tenderness, and juiciness. Seven represented the highest quality; one the lowest.

Shear values for a one-inch core from the anterior end of the cooked *pectoralis major* were determined on a Warner-Bratzler Shearing apparatus. Press fluid yields for a 25-g ground sample of the *pectoralis major* muscle were determined with a Carver Laboratory Press.

The *pectoralis major* muscle (anterior end) was used for nitrogen (Kjeldahl-Gunning-Arnold), fat (modified Babcock), moisture and ash determinations. Water soluble protein nitrogen was extracted with 0.06 M phosphate at pH 5.0 and heat-coagulable protein nitrogen removed after the sample was heated in a boiling water bath for 5 min.

RESULTS AND DISCUSSION

Roasting turkey to a satisfactory degree of doneness in all parts of the bird is difficult. The numerous muscles of the bird differ widely in size and shape and also in respect to the amount and distribution of fat, connective tissue, and myoglobin. For example, if the thigh is roasted to optimum doneness the breast may be less tender and underdone; whereas, if the breast is cooked satisfactorily, the thigh meat may be dry and stringy.

In this discussion the terms *pectoralis major* and *thigh* will be used to denote the muscle in which the end point temperature was taken. The terms *light* and *dark meat* refer to the samples removed from the birds irrespective of the muscle in which the end point of roasting was determined.

[a] The data are from theses by Katherine Cooley and Martine C. Ferguson in partial fulfillment of the requirements for the degrees of Master of Science.

[b] Contribution No. 212, Department of Home Economics, Kansas Agricultural Experiment Station, Kansas.

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Rate of heat penetration. Initial temperatures of the *pectoralis major* and thigh muscles were recorded just before the birds were placed in the oven and every 20 min thereafter (Figure 1). Initial temperatures of the muscles of the first year birds were slightly higher than those of the second year turkeys. Internal temperatures of first year birds of both muscles were similar for the first half of the roasting period; thereafter, the thigh temperatures were slightly higher than those of the *pectoralis major*. In the second year thigh temperatures were lower than those of the *pectoralis major* for the first 2 hr, but were higher for the remaining cooking time. When the birds were removed from the oven, internal temperatures of the thigh were higher than those in the *pectoralis major* in 65 of the 80 birds roasted. This agrees with Alexander, Schopmeyer and Lamb *(1)* who also reported that thigh temperatures were higher than those in the breast of well-done birds.

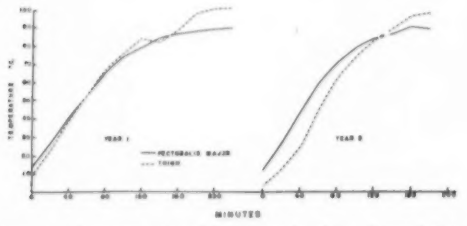

Figure 1. Average rate of heat penetration of roasted turkey halves.

Doneness. Tests for doneness included scoring tasted samples of light and dark meat and the appearance or color of the juice that exuded from the carcass as the birds were carved. In addition, the second year roasted birds were scored for doneness before being carved (Table 1). It was assumed that turkey should be cooked well-done, but not to the point of incurring excessive weight losses.

Data for all factors used to evaluate doneness, except scores for dark meat and juice in the second year, indicated significant differences among the turkey halves that could be attributed to the end point temperatures (Table 1). The first year birds cooked to an end point of 85° C in the thigh were the least done, as indicated by panel scores for light and dark meat and juice. Roasting turkey halves to 85° C in the *pectoralis major* and 90° C in the thigh gave similar results. These birds were considered done when judged by tasting, but slightly underdone when measured by appearance of the juice.

First year birds roasted to 90° C in the *pectoralis major* were more done, and were nearer the general concept of optimum doneness than those cooked to the other internal temperatures. Turkey halves cooked to 85° C in the thigh often were underdone; therefore, that end point temperature was eliminated during the second year.

In the second year, most of the criteria used for evaluating degree of doneness indicated that turkeys roasted to 90° C in the *pectoralis major* and 95° C in the thigh were more done than those cooked to end points of 85° C in the *pectoralis major* and 90° C in the thigh (Table 1). Light meat from birds cooked to 90° C in the thigh or in the *pectoralis major* and to 95° C in the thigh was scored about the same for doneness; meat from the birds given the two latter treatments was significantly ($P<.05$) more done than that from birds roasted to 85° C in the *pectoralis major*. When doneness was judged previous to carving, turkeys roasted to 85° C in the breast and 90° C in the thigh generally were similar and less done than those roasted to 90° C in the breast and 95° C in the thigh.

The size and uniformity of the *pectoralis major* muscle made it easier to place the thermometer in this muscle than in the thigh. Hence, an end point temperature of 90° C in the *pectoralis major* was preferred to 95° C in the thigh.

TABLE 1

Average doneness scores and cooking time with analyses of variance results and least significant differences

Factors	Significance of F values	Lsd[1]	85° C thigh	90° C thigh	85° C *pectoralis major*	90° C *pectoralis major*	95° C thigh
Doneness, 1st year[2]							
Light	*	0.2	2.7	3.2	3.0	3.5	....
Dark	*	0.2	2.7	3.0	3.1	3.4	....
Juice	*	0.4	2.0	2.7	2.4	3.2	....
Doneness, 2nd year[3]							
Light	*	0.6	....	4.3	3.9	4.5	4.8
Dark	ns	....	....	4.1	4.0	4.2	4.2
Juice	ns	....	....	3.8	3.5	3.8	3.7
Pricking drumstick	*	0.8	....	3.9	3.8	4.8	4.8
Pricking wing	*	0.8	....	3.9	3.9	4.6	4.8
Feel of drumstick	*	1.0	....	3.9	3.8	4.8	5.1
General appearance	**	0.8	....	4.0	3.7	5.0	4.8
Movement of joint	**	0.8	....	3.6	3.6	4.5	4.8
Cooking time							
(min/lb) 1st year	**	1.5	19.0	20.4	20.7	25.0	....
2nd year	***	2.7	....	21.8	20.7	25.7	27.1

[1] Least significant difference at the 5% level.
[2] A 5-point scale with a midpoint of 3 indicates optimum doneness; smaller number than 3 represent underdoneness and larger numbers, overdoneness.
[3] A 7-point scale with a midpoint of 4.
ns—nonsignificant.
*—significant at the 5% level.
**—significant at the 1% level.
***—significant at the 0.1% level.

Cooking time. Average cooking time, in min per lb, was greatest for the birds roasted to 95° C in the thigh, and just slightly less than that for birds roasted to 90° C in the *pectoralis major* (Table 1). Birds roasted to 85° C in the *pectoralis major* and to 85° and 90° C in the thigh averaged approximately the same time. The cooking time for birds roasted the first year to 90° C in the *pectoralis major* varied from 20.3 to 31.3 min per lb, whereas it ranged from 19.4 to 30.5 for second year turkeys.

Flavor, tenderness, and shear values. Flavor and tenderness were not affected by the end point temperatures to which the turkeys were roasted (Table 2). Average flavor scores for light and dark meat for birds given all treatments were similar within each year and between the 2 years of the study. Average tenderness scores for light meat were slightly higher than those for dark meat in both years.

Shear values for *pectoralis major* cores from the second year birds roasted to 95° C in the thigh were significantly ($P<.05$) lower than those from birds roasted to 90° C in the *pectoralis major* and 90° C in the thigh muscles. Correlation coefficients for shear values and tenderness scores for light meat ($r = -.327$, first year and $-.208$ second year) were nonsignificant.

Juiciness, press fluid yields and cooking losses. Juiciness scores for dark meat from the first year birds and those for the light meat from the second year turkeys were not affected significantly by the end point temperature (Table 2). However, significant differences attributable to the end point temperature were found among the juiciness scores for the first year light and second year dark meat samples. Light meat juiciness scores for first year birds roasted to 85° C in the thigh were significantly ($P<.05$) higher than for those roasted to 90° C in the *pectoralis major*. Juiciness scores for dark meat from second year birds roasted to 90° C in the thigh were significantly ($P<.05$) higher than those for dark meat from birds roasted to 85° C in the *pectoralis major* or 95° C in the thigh.

There were no significant differences among the press fluid yields from first year birds cooked to the various end points (Table 2). However, differences in press fluid yields attributable to the end point of roasting were very highly significant for the second year samples. Press fluid yields from birds roasted to 90° C in the thigh or 85° C in the *pectoralis major* were significantly ($P<.05$) higher than those from birds roasted to 90° C in the thigh.

Varying end point temperature had a highly or very highly significant effect on total, volatile, and dripping losses in all cases except for the dripping losses for the second year birds (Table 2). Turkeys roasted to 90° C in the *pectoralis major* and 95° C in the thigh averaged nearly 5% greater total cooking losses and these were significantly ($P<.05$) greater than for those cooked to 85° C in the *pectoralis major* and 90° C in the thigh. Total cooking losses for turkeys cooked to the same end points were similar for both years, but this was not true for volatile and dripping losses. Dripping losses were greater than the volatile losses for the first year birds and opposite results were obtained in the second year.

TABLE 2

Average palatability scores, shear values, press fluid yields and cooking losses with analyses of variance results and least significant differences

Factors	Significance of F values	Lsd[1]	85° C thigh	90° C thigh	85° C *pectoralis major*	90° C *pectoralis major*	95° C thigh
Flavor, 1st year							
Light	ns	....	5.4	5.6	5.5	5.5	....
Dark	ns	....	5.4	5.6	5.5	5.7	....
Flavor, 2nd year							
Light	ns	....	....	5.5	5.4	5.5	5.5
Dark	ns	....	....	5.3	5.2	5.5	5.2
Tenderness, 1st year							
Light	ns	....	5.8	5.9	5.8	6.1	....
Dark	ns	....	5.2	5.3	5.5	5.6	....
Tenderness, 2nd year							
Light	ns	....	....	6.1	6.3	6.4	6.6
Dark	ns	....	....	5.6	5.6	5.8	5.6
Shear value, 1st year	ns	....	11.6	11.9	12.9	12.6	....
Shear value, 2nd year	**	1.8	....	12.3	10.6	11.7	9.0
Juiciness, 1st year							
Light	*	0.3	5.2	4.8	4.9	4.4	....
Dark	ns	....	5.1	5.0	5.0	4.6	....
Juiciness, 2nd year							
Light	ns	....	....	5.3	5.0	4.8	4.8
Dark	*	0.5	....	5.7	5.2	5.3	5.0
Press fluid yields, 1st year	ns	....	9.6	8.9	9.5	8.8	....
Press fluid yields, 2nd year	***	0.5	....	9.8	9.6	8.8	9.1
Cooking losses, 1st year							
Total	**	1.8	16.4	19.3	20.0	24.1	....
Volatile	**	1.4	7.0	9.2	8.0	11.6	....
Dripping	**	1.1	9.3	10.1	12.0	12.6	....
Cooking losses, 2nd year							
Total	***	3.7	....	19.8	19.2	25.8	25.7
Volatile	**	3.1	....	14.0	12.8	17.9	18.4
Dripping	ns	....	....	5.7	6.2	7.7	7.1

[1] Least significant difference at the 5% level.
ns—nonsignificant.
*—significant at the 5% level.
**—significant at the 1% level.
***—significant at the 0.1% level.

TABLE 3

Summary of chemical analyses of pectoralis major samples, on wet basis

Factors	85° C				90° C			
	First year		Second year		First year		Second year	
	Raw	Cooked	Raw	Cooked	Raw	Cooked	Raw	Cooked
Total nitrogen, %	4.02	5.44	3.75	4.93	4.06	5.52	3.79	5.20
Water-soluble protein nitrogen[1]	30.08	11.1	28.3	12.6	31.1	11.0	29.8	12.7
Heat-coagulable protein nitrogen[1]	10.2	0.1	10.9	1.4	10.8	0.4	11.4	1.3
Fat, %	1.8	1.1	2.4	1.5	1.7	1.1	2.5	1.7
Moisture, %	72.1	63.1	73.9	66.5	72.1	62.9	73.3	64.3
Ash, %	1.07	1.10	0.98	1.06	1.07	1.09	1.02	1.09

[1] % of total nitrogen.

On the whole, turkey halves roasted to 90° C in the *pectoralis major* and 95° C in the thigh had lower juiciness scores and press fluid yields, but greater cooking losses than those cooked to 85° C in the *pectoralis major*, 85° C in the thigh and 90° C in the thigh.

Chemical analyses. It was postulated that the heat-coagulable protein nitrogen in the cooked meat might be related to the doneness of the birds. Although there were differences in the degree of doneness as measured subjectively, no relationship between these and heat-coagulable protein nitrogen were found (Table 3). Also, appreciable difference in moisture was noted that could be attributed to end point temperatures. Slightly less fat was present in the cooked *pectoralis major* than in the raw, even though moisture was lost during roasting.

SUMMARY

Eighty frozen defrosted Broad Breasted Bronze turkey halves were roasted at 163° C (325° F) to end point temperatures of 85° and 90° C in the *pectoralis major* and 85°, 90° and 95° C in the thigh.

Turkey halves roasted to either 90° C in the *pectoralis major* or 95° C in the thigh were satisfactorily done. However, an end point temperature of 90° C in the *pectoralis major* was preferred to 95° C in the thigh because of the size and greater uniformity of the *pectoralis major* muscle. Turkeys roasted to 85° C in the *pectoralis major* and 90° C in the thigh were similar and less done than those roasted to 90° C in the *pectoralis major* or 95° C in the thigh. Turkey halves cooked to end point temperatures of 85° C in the thigh were underdone.

Flavor and tenderness scores were similar for birds cooked to all end point temperatures. Juiciness scores and press fluid yields were lower and cooking losses higher for satisfactorily done than for underdone turkey halves. The amount of heat-coagulable protein in the *pectoralis major* was not related to the doneness of the muscle.

Acknowledgment

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Studies on Domestic Dates. I.

Methods for Evaluating Darkening

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STUDIES UNDERWAY in this laboratory on the chemistry of deterioration of domestic dates necessitated the development of reliable objective methods for measuring their darkness. Such methods make possible the quantitative determination of the effects of variables such as temperature, moisture content and oxygen concentration on the rate of darkening.

It was considered essential in the present work to be able to measure darkening in terms of both the observable darkness and the dark pigment content of date tissue because of the expected complexity of the browning reactions involved *(6, 7, 9, 10, 13)*. Solvent extraction methods in which the soluble pigments are extracted from the darkened tissue and their concentrations determined colorimetrically are useful for measuring dark pigment concentration *(2, 4, 8, 11)* but the results are not necessarily related to or convertible into units of observed darkness. Reflectance measurements, on the other hand, are closely related to the visually observed color of the sample *(1, 5, 12)*, but do not measure the dark pigment concentration. Lack of a single procedure by which observable darkness and the dark pigment concentration could be determined simultaneously made it necessary for our purposes to develop both a reflectance and a solvent extraction method for measuring the darkness of dates.

Objectives of the work reported in this paper therefore were: (1) to develop a reflectance method for measuring the darkness of dates; (2) to develop a solvent extraction method for measuring the amount of dark pigment produced in darkened dates; and (3) to evaluate and compare both methods over a wide range of darkness values using identical samples of dates.

SOLUBLE PIGMENT EXTRACTION METHOD

Extraction procedure. Numerous tests showed that blending 20 g of ground (food chopper) date tissue with 100 ml of 65% (v/v) aqueous methanol for 3 min followed by filtration through S & S No. 597 filter paper, washing with three 45 ml portions of fresh solvent, addition of 2 ml of 4 M acetate-citrate buffer to adjust the pH to 6.0 to 6.3 and final dilution to 250 ml was the best extraction procedure. Pigment concentration was determined by measuring the absorbance at 400 mμ with a DU spectrophotometer. Ninety-six per cent of the pigment which could be removed from dark date tissue by repeated extractions with this solvent was obtained using this procedure. Tests with several other pigment extracting solvents *(8)* and various methanol-water mixtures showed that 65% methanol was the most suitable solvent. Increasing the blending time to 5 or 10 min gave no significant increase in extractable pigment concentration. Clear solutions were obtained with one filtration. Allowing the unbuffered extract to stand overnight at room temperature produced no change in absorbance at 400 mμ.

Pigment concentrate. Absorption spectra of extracts from light and dark date tissue are given in Figure 1. They are very similar except for the difference in pigment concentration. Extracts of dark dates were treated to obtain a concentrate of the dark pigment. The colored material was precipitated with lead acetate, washed thoroughly with water, suspended in methonal, and treated with hydrogen sulfide. The lead sulfide precipitate was removed under vacuum at 50° C and the pigment concentrate obtained as a dark-brown, heavy syrup. The absorption spectrum of this material was very similar to that of the crude extracts. It differed only in having a somewhat higher absorption in the 450-600 mμ region. Dilutions of the pigment concentrate with 65% methanol adjusted to pH 6.2 with acetate-citrate buffer showed that Beer's Law was followed up to a concentration of 1.2 mg/ml

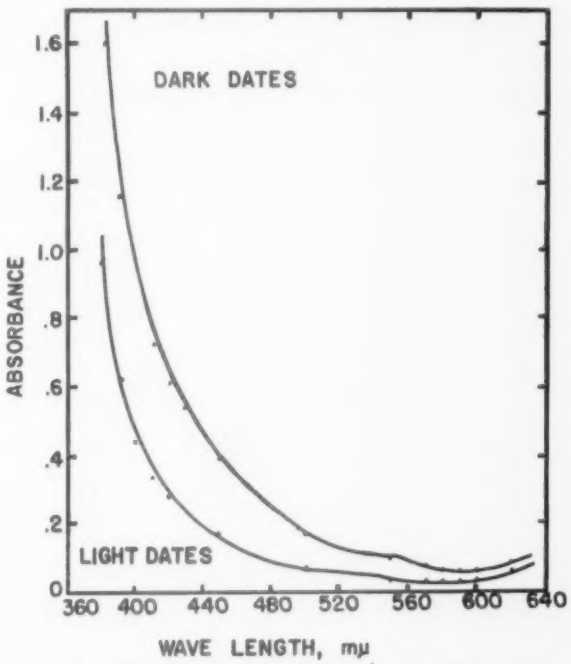

Figure 1. Absorption spectra of 65% aqueous methanolic extracts of light and dark dates.

[a] Collaborator employed by Date Administrative Committee, Indio, California, with whom this work was conducted cooperatively.

[b] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture, Albany 10, California.

TABLE 1
Estimate of the precision of methods used in determining darkness

Method	Replicates	Light dates			Dark dates		
		Range	Standard deviation	Coefficient of variation	Range	Standard deviation	Coefficient of variation
Extraction, mg pigment/g dry tissue	6	4.57– 4.74	.0611	1.32%	8.91–9.20	.151	1.67%
Reflectance, %, whole dates	15	4.35– 5.20	.278	5.88%			
Reflectance, %, pureed dates	6	18.1 –19.3	.535	2.86%	4.40–4.78	.175	3.90%

at 400 mmc. It gave a specific extinction coefficient (k) of 0.650 ml/(mg)(cm). By use of this extinction coefficient the absorbance readings at 400 mmc can be converted to mg dark pigment/ml of extract. If the moisture content of the tissue is known mg pigment/g dry tissue can then be calculated.

Test of precision. The precision of the method was tested by use of six replicates each of light and moderately dark date tissue. The standard deviations expressed as a percentage of the mean pigment concentration were less than 2.0 in each case (Table 1).

REFLECTANCE METHODS

General instrumentation. A Photovolt photoelectric reflection meter[c] equipped with an external constant voltage transformer and galvanometer was used in this work. The search unit was equipped with blue, green, and amber tristimulus filters. The instrument was usually operated with suppressed zero to obtain the widest possible spread of readings between light and dark dates. The 100 value was set by use of enamel plates (standardized by the instrument manufacturer against a freshly prepared magnesium oxide surface representing 100% reflectance) whose reflectances with the green filter ranged from 36.5 to 8.0%. The zero value was set by use of a dark cavity whose reflectance was zero or by a black enamel plate which had a reflectance of 1.5%. The instrument was standardized before each reading by placing the working standards in the bottom of the plastic sample dish in a reproducible manner. This procedure cancelled out any differences between sample dishes. The green tristimulus filter was always used, except in the case where other filters are specifically mentioned.

To increase the speed and effectiveness of reflectance measurements a special sample presentation device was added to the reflectometer. This device permitted the average reflectance value of a large sample to be obtained in one operation through the use of a rotating plastic sample holder. Other workers have shown this method to be equivalent to the average of a large number of separate determinations *(8, 10)*. This was also true in the present work.

The rotating sample presentation device used was patterned after that of Bockian and Hirzel *(1)*. The scanning unit of the reflection meter is contained in the box shown in Figure 2. The unit is attached to a sliding mount which allows it to be moved so that most of the bottom surface of the sample dish can be scanned from three positions. The dish is supported 1/32 in above the scanning unit by three vertically-positioned wheels and is kept in place while rotating by three horizontally-positioned side wheels. One set of wheels is fixed to a moveable, spring-loaded mount so that the sample dish can be positioned or removed without difficulty. The sample dish is rotated by a variable-speed motor acting through a friction-drive wheel kept in contact with the dish by spring tension. The sample container is a circular, styrene dish 5 1/16 inches in diameter, 1 1/2 in high with a perfectly flat 1/16 in thick bottom. The speed at which the sample dish was rotated was generally between 30 and 60 rpm depending on the homogeneity of the sample. The more homogeneous the sample the slower the speed of rotation necessary to give a constant galvanometer reading.

Each sample dish was tested initially for uniformity by measuring its reflectance when filled with a homogeneous celite-charcoal mixture which had a reflectance similar to that of light-colored dates. The equivalency of the three scanning unit positions was tested in a similar manner and found to be identical. In addition, the average of adjacent static readings was compared with single readings taken when the dish was rotating. No significant difference was observed. The theory supporting this observation was discussed by Lukens and Creese *(3)*.

The change in color of domestic Deglet Noor dates from golden-brown to dark-brown to black is due primarily to a decrease in the lightness attribute of color. In tristimulus colorimetry the green filter is used to directly measure lightness or luminous apparent reflectance (Y value). This filter was selected, therefore, since it was expected to give the best single measure of the color change. The assumption was substantiated by measurements in which the blue, green and amber tristimulus filters were used to calculate the X, Y, Z tristimulus values. In X, Y and Z values, the light and dark dates differed by 0.90, 2.30 and 0.023 respectively.

Figure 2. Sample dish and presentation device for reflection meter.

Whole dates. In order to obtain a reasonably uniform surface dates were carefully pitted, placed in the sample dish and an aluminum disk of slightly smaller diameter than the sample dish was placed on the dates and pressure applied with a hydraulic press. It was convenient to leave the aluminum disk in place during the reflectance reading since its removal caused the dates to separate from the bottom of the dish. A pressure of 200 psi was found to give a uniform surface without rupturing the skins of dates containing 20% moisture. Optimum pressures for dates of widely different moistures would probably vary somewhat from this figure. The effect of successive increases in pressure on the reflectance readings is shown in Table 2. When a pressure of 200 psi was applied to the same sample three successive times identical values were obtained showing that the decrease in reflectance is due to the increased pressure.

The reproducibility of this method was tested using a large lot of dates which were visually selected as being as closely equivalent in color as possible. A weighted average method based on the percentage of the total area scanned in each position was used to obtain the overall reflectance value. For

[c] The mention of special instruments or materials does not imply endorsement by the Department of Agriculture over others of similar nature.

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TABLE 2

Effect of pressure on reflectance of whole dates

Pressure [1] psi	% Reflectance	
	Sample 1	Sample 2
100	2.95	3.14
150	2.57	2.51
200	2.52	2.41
250	2.44	2.36

[1] Pressure maintained for 5 sec in successive steps.

scanning unit positions 1, 2 and 3 these areas were 11.1, 36.5 and 52.4%, respectively, of the total area. The reproducibility, Table 1, is fair as shown by the standard deviation obtained, i.e., 5.9% of the mean reflectance.

Pureed dates. Pitted date tissue (120 g) was blended with 1.5 times its weight of water. This amount was just sufficient to produce a heavy puree which could be transferred without difficulty. Small variations (±3%) in the amount of water, which are equivalent to large changes (10–30%) in the moisture content of the date tissue, caused only a ±1.5% change in the reflectance values obtained. The puree (250 g) was poured into the sample dish and its reflectance determined; the rotating dish method was used. It was necessary to protect the instrument from outside light by using a covered sample dish which was painted black on the sides and top. A 10% variation in the amount of puree put in the sample dish caused no significant difference in the reflectance readings.

The reproducibility of this method was tested. Large amounts of material were prepared from light and dark dates of equivalent moisture contents by removing the pits and passing the tissue through a food chopper several times. One hundred and twenty gram portions were then pureed with water and the reflectance of each was measured. The standard deviations (Table 1), 2.86% and 3.90% of the mean values for light and dark tissue, are considerably smaller than that obtained with whole dates. The range of reflectance values between light and dark dates is also much wider when the puree method is used.

COMPARISON OF EXTRACTION AND REFLECTANCE METHODS

Large quantities of homogeneous light and dark tissue were prepared as before from light and dark dates. Various mixtures, by weight, of the light and dark tissue were made and the soluble dark pigment content and reflectance (puree) of each determined. The data in Figure 3 show that the amount of soluble dark pigment extracted is directly related to the percentage of dark tissue present. Thus, pigment extraction is uniform for all levels of tissue darkness tested. Reflectance of the puree, as would be expected,

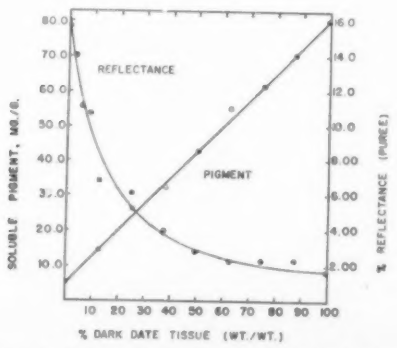

Figure 3. Reflectance (puree) and soluble dark pigment content of mixtures of light and dark date tissue.

is not directly related to the percentage of dark tissue in the mixture, Figure 3; in fact it approaches a logarithmic relationship. It is apparent from Figure 3 that the reflectance is extremely sensitive to the darkening of light colored dates, but is relatively insensitive to the darkening of dates which have a reflectance of less than 3%.

The methods were also compared by making use of dates of different darkness levels. Ten batches of dates ranging in darkness from extremely light (SO_2 bleached) to very dark (prolonged high temperature storage) were prepared and arranged in order of increasing darkness (Nos. 1–10) by visual inspection. The dates within each group were selected for maximum color uniformity. The darkness of each batch was then determined using each of the three methods: extractable dark pigment, reflectance of whole dates and reflectance of pureed dates.

The results of this study, Table 3, show the range of values to be expected from extremely light and dark

TABLE 3

Comparison of pigment extraction and reflectance methods for determining darkness of dates

Sample No.	Treatment	Whole dates, % reflectance	Pureed dates, % reflectance	Extracted dates, soluble pigment mg/g dry wt
1	SO_2 bleached	10.1	32.9	5.03
2	SO_2 bleached	8.89	27.5	5.14
3	None	6.74	23.7	5.83
4	None	3.47	22.2	6.39
5	Stored at 60° C	2.78	13.0	8.12
6	Stored at 60° C	1.20	7.26	16.4
7	Stored at 60° C	1.07	6.12	17.0
8	Stored at 60° C	0.75	4.21	28.0
9	Stored at 60° C	1.09	3.82	29.8
10	Stored at 60° C	0.85	2.68	50.6

dates. The three-fold greater reflectance range exhibited by the pureed dates as compared with the whole dates is caused by the dilution effect of the water and the higher reflectance of the more uniform surface of the puree. Except in the very low reflectance region where sensitivity is poor an approximate linear relationship exists between the reflectance values obtained using these two methods. Thus, the reflectance of pureed dates can be used as an estimate of the reflectance of whole dates. In addition, the reflectance values compared very well with the visually observed darkness of the dates.

The amount of dark pigment extracted from dates of different darkness levels, Table 3, is not directly related to the observed darkness as measured by reflectance. This agrees with the relationship between reflectance and soluble pigment shown in Figure 3. Table 3 also shows that the usefulness of the extraction method is not limited by the darkness level of the dates as is the reflectance method. Below 3% reflectance extractable dark pigment is a much more reliable measure of darkening.

Samples of these dates were used, in addition, to obtain an estimate of the darkness level which limits their acceptability. Judges were asked to choose the sample of maximum darkness that they would purchase if buying dates for home use. Of the 16 judges 81% chose samples No. 5–7 while 19% picked samples

No. 8–10 (sample numbers are shown in Table 3). When asked their preference with regard to darkness most of the judges (87%) chose samples No. 3 and 4. Close examination of the dates revealed that sample No. 7 still retained a slight translucency characteristic of light dates whereas samples No. 8–10 had none. This translucency is caused by the lighter color of the fibrous sheath underlying the darker, soft outer tissue. When this sheath becomes equally as dark as the outer tissue the translucency disappears. Thus, the darkness level associated with sample No. 7, approximately 6% reflectance for pureed dates, can logically be employed as a useful experimental criterion of color deterioration in dates.

SUMMARY

Two methods have been developed by which the darkening of dates during deterioration can be followed. The reflectance method in which either whole or pureed dates can be used is very sensitive to the initial darkening and is closely related to the visually observed darkness. The use of pureed dates is preferred, however, because of the greater precision and the broader range of reflectance values obtained with light and dark dates. The solvent extraction method permits the precise measurement of the amount of soluble dark pigment present in date tissue of any degree of darkness. Although the results are not directly related to the visually observed darkness this method has the advantage of providing a measure of the later stages of darkening where the reflectance method is not sensitive.

Acknowledgments

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The Use of Antibiotics and Gamma Irradiation in the Aging of Steaks at High Temperatures[a, b, c]

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IN THE MEAT INDUSTRY, the aging of beef carcasses and cuts to improve tenderness is frequently carried out by holding the meat for two weeks at 35° to 40° F. This procedure requires large inventories and results in moisture and trimming losses. For these reasons beef usually moves directly to the retail outlet without aging. A rapid, economical method of aging would permit more beef, particularly the lower grades, to be tenderized in this manner.

Assuming that the tenderization which occurs during aging is an enzymatic process, it follows that the aging time can be reduced if the temperature is raised. Weiser *et al (9)* aged beef rounds and carcasses at room temperature (75–85° F) after infusion with an antibiotic solution and found them to be more tender than controls which had been chilled after slaughter. In other experiments aging beef for 2–3 days at temperatures ranging from 57° to 86° F, produced tenderness changes similar to those found in beef aged 12–14 days at 35° to 40° F *(6, 7)*. In this work ultraviolet irradiation was used to control surface microbial growth.

In the present study tetracycline antibiotics alone and in combination with gamma radiation were used as preservatives during aging at elevated temperatures. Niven and Chesbro *(4)* have reported that high energy irradiation and the tetracycline antibiotics complement each other in their preservative effect on meat. The adverse effects of irradiation on meat flavor are generally proportional to the dosage employed but are not pronounced at "pasteurization" levels. Beef has been shown to be more susceptible than pork to flavor and odor alteration *(3, 5)*. In the present study, both antibiotics and irradiation were used, when needed, to allow maximum flexibility in holding conditions with the objective of establishing the optimum aging time and temperature for the rapid tenderization of beef.

EXPERIMENTAL METHODS

Preliminary work was conducted in which the experimental procedures and the level of irradiation were established. In combination with antibiotic treatment 45,000 to 50,000 rad of gamma radiation was found to be sufficient to control microbial growth under the conditions employed. Although irradiation at higher levels was somewhat more effective, dosages of 100,000 rad or more resulted in detectable flavor changes in the aged steaks.

Inside rounds from chilled U. S. Good and Utility beef carcasses, obtained approximately 72 hr post slaughter, were used in these studies. The rounds were stitch pumped to 106% of their weight with a solution containing an appropriate amount of antibiotic. The stitch pumping procedure was found to be effective when 30 to 50 ppm of antibiotic were infused into the meat prior to aging. It was assumed that the other methods which would provide completely uniform distribution would permit a lower level of antibiotic to be employed.

The *semimembranosus* muscle from the rounds was sliced into ¾ in steaks and allocated to the aging conditions in a randomized design so that all steak locations, left and right sides and carcasses were, as far as possible, represented in all treatments of a particular experiment. The beef was aged in the form of steaks which allowed adjacent locations within a muscle to be subjected to different treatments. This procedure was chosen because it most nearly approached using identical samples for two treatments and, in addition, took advantage of the similarity between sides. Previous work *(1)* in this laboratory had shown that steaks 4–9 (anterior to posterior) were the most uniform in tenderness and these were used in the aging experiments.

A surface dressing on each steak was obtained by placing the steak in a plastic bag (Cry-O-Vac)[e] containing 50–100 ml of pumping solution. The bag was agitated, allowed to stand for 30 min and the fluid content decanted prior to sealing the package under vacuum. All preparation work was conducted at 45° F.

One experiment was designed to determine whether irradiation would interfere with tenderization at elevated aging temperatures. In this instance the packaged steaks were held overnight at 35° F and then transported to the Argonne National Laboratory in a refrigerated container. Irridiation was performed in a measured gamma field obtained from spent fuel rods. Refrigeration sufficient to maintain a steak temperature of 35° to 40° F during irradiation was provided by refrigeration plates. This preparatory work was completed in approximately 30 hr.

Aging at 90°, 100°, 110° and 120° F was conducted in convection type incubators, all steaks being at the same height in the incubator. For aging at 35° F, a forced air cooler was used. Unaged control steaks were frozen at −20° F immediately following cutting.

A bacteriological examination of the steaks was made after aging. A weighed amount of meat was removed aseptically from a steak and homogenized in a Waring blender in sterile distilled water. Aerobic counts were made by the conventional quantitative plating technique using tryptone-yeast extract-glucose

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[a] Journal Paper No. 194, American Meat Institute Foundation.

[b] This paper is a report of work done under contract with the U. S. Department of Agriculture and authorized under the Research and Marketing Act of 1946. The contract was supervised by the Eastern Utilization Research and Development Division of the Agricultural Research Service.

[c] Presented at the Eighteenth Annual Meeting of the Institute of Food Technologists, Chicago, May 27, 1958.

[d] Present address, Microbiology Department, University of New Hampshire, Durham, N. H.

[e] Supplied by the CRYOVAC Company.

agar *(2)*, incubated for 48 hr at 86° F (30° C). Populations of anaerobic bacteria were estimated by adding 0.1 ml of a suitable dilution of the meat infusion to 20 ml of medium in a screw cap tube. Brewer's medium (Difco), modified to contain 0.1% soluble starch was employed. The cultures were incubated for 3 days at 86° F (30° C). Although it was realized that this technique lacked precision because of growth by facultative bacteria, it served the purpose for which it was intended in these experiments.

A further test for the presence of toxic substances was made by inoculating mice intraperitoneally with 0.2 ml of the filtrate from the water and meat homogenate used for plate counts.

After the samples for bacteriological analysis were removed, the steaks were wrapped in aluminum foil and frozen. When the bacteriological examination was completed the steaks were thawed overnight at 45° F prior to broiling. Cooking and sampling procedures were the same as described in earlier reports *(1, 8)*. Initial tenderness of the steaks and the amount of residue remaining after chewing were recorded using a scoring range from 1–10 corresponding to increasing tenderness and decreasing residue. Panel data were analyzed by analysis of variance or Student's "t" test. For shear measurements, 5–7 cores, ½ in in diameter, were cut from the portion of the cooked steaks not used for the taste panel. The cores were brought to 45° F and sheared perpendicular to the fibers using a Warner-Bratzler shear machine.

Specific procedures applying to individual experiments are given prior to the results and discussion of each.

RESULTS AND DISCUSSION

Effect of gamma irradiation on tenderness. In a preliminary experiment, irradiation in excess of 100,000 rad had an apparent adverse effect on the tenderness of the steaks during subsequent aging. The following experiment was designed therefore, to determine whether irradiation at 45.5 thousand rad had a significant direct effect on the tenderness of steaks or on tenderization at elevated temperatures. An aging temperature of 110° F was used. Matched inside rounds of 8 U. S. Good carcasses were infused with oxytetracycline,[f] 2 pairs with 20 ppm and 6 pairs with 30 ppm. After cutting into steaks and packaging, one half of the steaks were irradiated. One half of the steaks from the irradiated group and one half from the non-irradiated group were randomly allocated to an aging temperature of 110° F for 24 hr and the remainder were frozen immediately as controls.

Microbiological control of both irradiated and non-irradiated steaks was generally satisfactory. Aerobic counts averaged less than 100,000 organisms per g and the anaerobic counts were usually lower than the aerobic counts. The irradiated steaks had slightly lower aerobic and anaerobic counts but the antibiotic treatment alone yielded satisfactory control during aging and was used subsequently when aging was carried out at 110° F. The relative effectiveness of gamma irradiation was more pronounced in subsequent experiments when the temperature was more nearly optimal for bacterial growth.

The mean tenderness and residue scores, shear values, and their statistical analyses are given in Tables 1a and 1b respectively.

Taste panel results confirmed preliminary observations that irradiation of steaks at a level of 45,500 rad had no adverse effect on the flavor of the product. This level of irradiation did not affect the tenderness of unaged steaks or inhibit tenderization when steaks were subsequently aged at 110° F. Significant increases in tenderness were found in steaks aged at 110° F for 24 hr.

[f] Supplied by Chas. Pfizer and Co., Inc.

TABLE 1a

Tenderness values of steaks receiving 45.5 thousand rad of gamma radiation prior to aging at 110° F

Treatment	Hours (at 110° F)	No. of judgments	Tenderness		Shear[1] (lb)
			Initial	Residue	
Control	0	180	5.3	5.9	10.1
Control	24	183	6.6	6.7	9.2
Irradiated	0	185	5.4	6.0	10.1
Irradiated	24	185	6.4	6.6	9.4

[1] Mean of 24 steaks, 5–7 determinations per steak.

Aging at 90° F. Matched inside rounds from 8 U. S. Good carcasses were infused with oxytetracycline (OTC). The first 2 pairs received 30 ppm OTC but because microbial control was not entirely satisfactory, the remaining 6 pairs received 50 ppm OTC. After packaging, the steaks received approximately 45,500 rad of gamma radiation and were aged 0, 1, 2, or 3 days at either 35° or 90° F.

TABLE 1b

Analysis of variance for tenderness and residue scores and shear values of irradiated steaks aged at 110° F

Source of variation	df	Tenderness		
		Initial ms	Residue ms	Shear ms
Irradiation	1	0.03	0.79	0.97
Animal	7	0.88	1.02	16.51***
Aging	1	29.48***	16.42***	17.10***
Residual	86	0.80	1.43	2.25

*** $P<0.001$.
df—degrees of freedom.
ms—mean square.

Microbial growth was satisfactorily controlled in this experiment. The anaerobic counts ranged from 1×10^3 to 240×10^3 organisms per g of meat depending on the aging time.

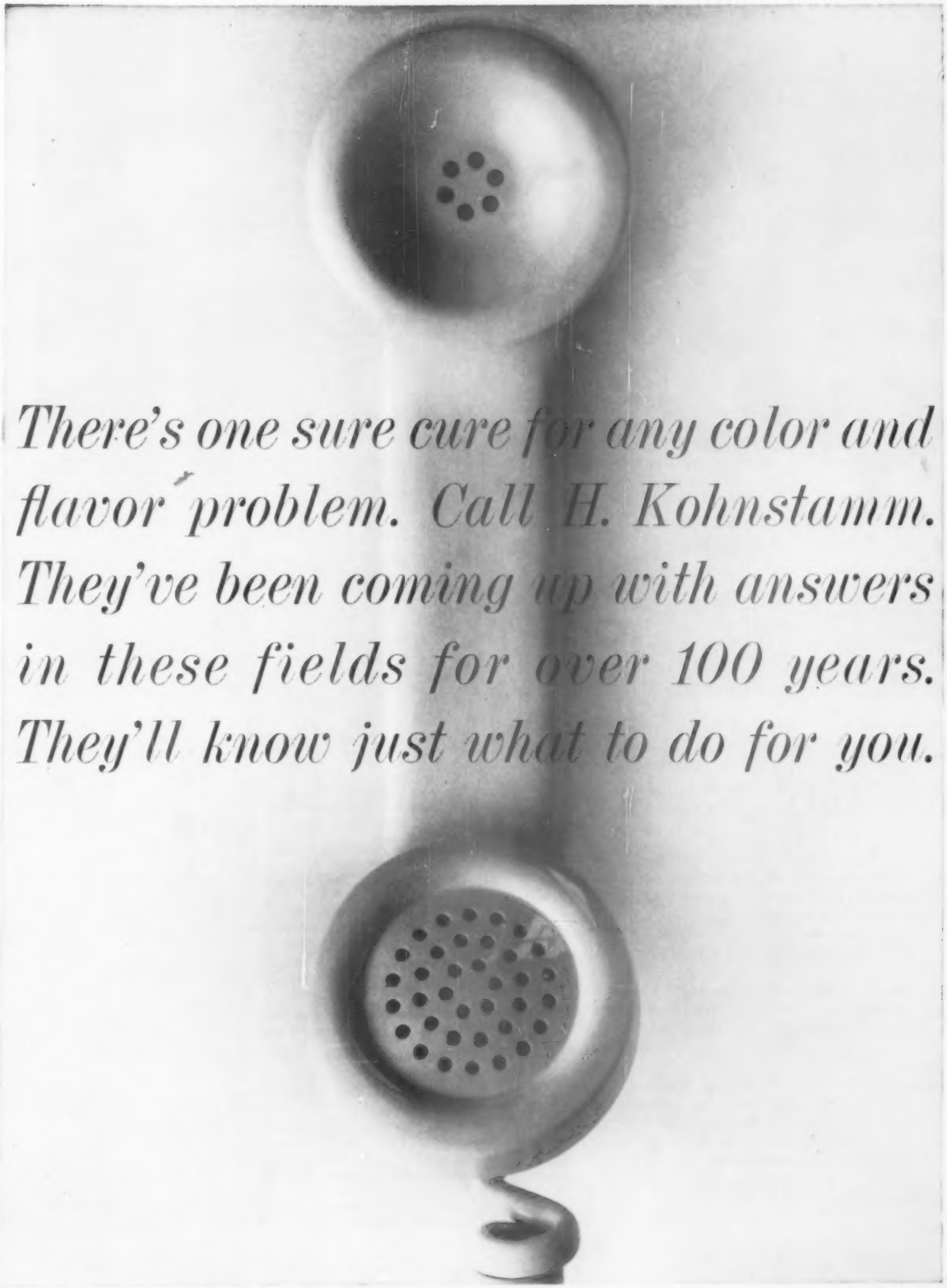
The tenderness and residue scores, shear values, and the analysis of the results are shown in Tables 2a and 2b, respectively. Steaks held at 90° F for 1, 2, or 3 days had significantly higher tenderness and residue scores than unaged controls. Some increases in tenderness were noted in the steaks held at 35° F but in

TABLE 2a

Effect of time and temperature of aging on the tenderness scores and shear values of semimembranosus steaks

Days aged	Tenderness[1]				Shear values[2]	
	Initial		Residue			
	35° F	90° F	35° F	90° F	35° F	90° F
					(lb)	(lb)
0	5.5	5.4	5.6	5.5	10.1	9.9
1	6.0	6.0	6.1	6.2	9.9	9.7
2	5.7	6.5	5.6	6.5	10.4	8.7
3	5.9	6.2	5.8	6.2	9.7	9.3

[1] Mean of 96 judgments for each time-temperature group.
[2] Mean of 12 steaks, 5–7 values per steak.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Ad No. 5-60

TABLE 2b

Analysis of tenderness scores and shear values of steaks aged at 90° F

Source of variation	df	Tenderness		Shear	
		Initial ms	Residue ms	df	ms
Time	3	15.17**	13.11**	3	8.60
Temp	1	9.99	15.05*	1	66.17*
T x T	3	11.04**	9.50*	3	19.21
Residual	756	2.84	2.83	625	10.27
L.S.D. .05		.50	.49		1.00
L.S.D. .01		.64	.64		1.30

* $P<0.05$.
** $P<0.01$.

general these differences were not significant. Differences in shear values were significant only for temperature effect. Because temperature had a significant effect on residue scores and not on initial tenderness scores may indicate that the Warner-Bratzler shear is a better measure of changes in connective tissue than of changes in muscle fibers during aging. The greatest increase in tenderness, as determined by the three measurements of tenderness, was in those steaks which were held for 2 days at 90° F. It will be observed in Table 2a that the steaks reached an apparent optimum tenderness after 2 days. A similar effect occurred at other aging temperatures and is discussed in conjunction with the experiment which follows.

Aging at 110° F. In this experiment, 4 pairs each of U. S. Good and Utility inside rounds were infused with 50 ppm of oxytetracycline and prepared for aging as previously outlined. U. S. Good rounds were used in the second half of this experiment because it was thought that any aging effects in the steaks from Utility grade rounds were being partially masked by the large amount of connective tissue noted by the panel members. The steaks were aged 0, 16, 24 or 40 hr at 35° to 110° F.

With the exception of the steaks which were aged 40 hr at 110° F the bacteria counts on samples of the aged product were generally low. Six of the 12 steaks aged for 40 hours at 110° F had apparent anaerobic counts in excess of 100,000 organisms per g and were positive for staphylococci. Taste panel judgments were not made on these steaks.

The taste panel and shear test results on the steaks from U. S. Good and Utility rounds are shown in Table 3a. The analysis of these data appears in Table 3b. The tenderness of the steaks aged at 110° F increased significantly at all aging times. The steaks which showed the greatest difference in tenderness compared to the controls held at 35° F were those which were aged 24 hr at 110° F. These differences were apparent in tenderness and residue scores as well as shear test values. A marked difference in taste panel scores and shear values was obtained between grades but the extent of tenderization within each of the grades was quite comparable. As indicated previously, aging of steaks in this manner produced an apparent optimum tenderization followed by some loss of tenderness.

In this experiment evaporation losses during storage, defrosting, and cooking were noted. Moisture losses from the steaks aged 40 hr at 35° and 110° F averaged 6.2 and 17.6%, respectively. These losses were approximately 22 and 7% greater than occurred after 24 hr aging. Cooking losses were less in those steaks which were aged at 110° F but the total losses were 8% higher in steaks aged for 40 hr than they were for steaks aged for 24 hr. It is felt that the greater moisture loss in the steaks aged for 40 hr at 110° F was at least in part responsible for the lower tenderness scores of these steaks,. Weight losses in these experiments could be expected to be higher than under conditions of normal aging because of the moisture added in the antibiotic treatment.

Bacterial spoilage was more conveniently controlled at 110° F than at 90° F. In addition, greater tenderization was noted when steaks were aged at the higher temperature and shorter time. For these reasons 110° F was accepted as being more nearly the optimum for aging and a further experiment was de-

TABLE 3b

Analysis of variance of taste panel scores and shear values of steaks aged at 35° F and 110° F

Source of variation	df	Tenderness		Shear	
		Initial ms	Residue ms	df	ms
Temperature	1	286.3***	36.6***	1	93.2***
Time	3	84.0***	5.3*	3	73.1***
Grade	1	113.0***	34.6***	1	5164.2***
Residual	636	2.4	2.47	512	10.24

* $P>0.05$.
*** $P>0.001$.

TABLE 3a

Mean tenderness scores and shear values of steaks from Utility and Good rounds aged at 110° F

Grade	Aging time	No. of judgments	Tenderness				Shear values	
			Initial		Residue		35° F	110° F
			35° F	110° F	35° F	110° F		
	(hr)						*(lb)*	*(lb)*
Utility	0	42	4.6	5.7	5.2	6.2	14.7	14.5
	16	42	4.9	6.2	5.7	6.6	16.2	13.2
	24	47	5.3	7.4	5.6	7.1	13.7	12.5
	40	22	4.7	5.7	4.8	6.0	15.6	13.2
Good	0	48	5.9	5.9	6.4	6.5	7.6	7.7
	16	48	6.3	6.9	6.4	7.0	8.7	7.8
	24	48	6.5	7.5	6.8	7.2	8.1	7.1
	40	24	5.9	6.7	6.5	6.9	7.7	7.7

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TABLE 2b

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Source of variation	df	Tenderness		Shear	
		Initial ms	Residue ms	df	ms
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Temp	1	9.99	15.05*	1	66.17*
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Mean tenderness scores and shear values of steaks from Utility and Good rounds aged at 110° F

Grade	Aging time	No. of judgments	Tenderness				Shear values	
			Initial		Residue		35° F	110° F
			35° F	110° F	35° F	110° F		
	(hr)						(lb)	(lb)
Utility	0	42	4.6	5.7	5.2	6.2	14.7	14.5
	16	42	4.9	6.2	5.7	6.6	16.2	13.2
	24	47	5.3	7.4	5.6	7.1	13.7	12.5
	40	22	4.7	5.7	4.8	6.0	15.6	13.2
Good	0	48	5.9	5.9	6.4	6.5	7.6	7.7
	16	48	6.3	6.9	6.4	7.0	8.7	7.8
	24	48	6.5	7.5	6.8	7.2	8.1	7.1
	40	24	5.9	6.7	6.5	6.9	7.7	7.7

signed to investigate aging temperatures within a 10° F range of the apparent optimum.

Aging at 100° F, 110° F and 120° F. Matched pairs of the inside rounds of six U. S. Good carcasses containing 50 ppm oxytetracycline were sliced and the steaks packaged, aged and stored according to the procedures previously described. Steaks aged at 100° F were held 2 days and those aged at 110° F and 120° F were held for 24 hr. Control samples were frozen immediately after packaging or after irradiation. The steaks that were aged at 100° F and their adjacent controls received 45.5 thousand rad of gamma radiation prior to aging. Bacterial growth was effectively controlled and all the steaks were submitted to the taste panel. Taste panel results and shear test scores are shown in Table 4. The differences between mean values of the various experimental treatments were analyzed by the "t" test and significant differences are indicated in the table.

As indicated in Table 4 inital tenderness and residue scores were related directly to the aging temperatures and were increased significantly over comparable unaged controls at all temperatures. In the lower portion of Table 4 comparisons are drawn between three aging conditions. The difference between mean scores for steaks aged at 100° F and 110° F are not significant and are in both instances significantly lower than those of steaks aged at 120° F. Although

TABLE 4

Mean tenderness, residue and shear values for steaks aged at 100°, 110°, and 120° F

Time and temp. of aging	No. of judgments	Tenderness		Shear	
		Initial	Residue	No.	(lb)
2 days at 100° F	114	6.0	6.1	77	8.75
Unaged controls	114	5.3**	5.5**	69	9.58NS
1 day at 110° F	112	6.3	6.2	84	8.69
Unaged control	112	5.1	5.2	89	9.73
1 day at 120° F	106	7.4	7.2	73	7.04
Unaged control	106	5.2***	5.4***	83	9.50***
2 days at 100° F	114	6.0	6.1	77	8.75
1 day at 110° F	112	6.3	6.2	84	8.69NS
2 days at 100° F	114	6.0	6.1	77	8.75
1 day at 120° F	106	7.4***	7.2	73	7.04***
1 day at 110° F	112	6.3	6.2	84	8.69
1 day at 120° F	106	7.4***	7.2***	73	7.04***

* P>0.05.
** P>0.01.
*** P>0.001.

the scores alone showed that 120° F was the preferred aging temperature, steaks aged in this manner were reported by the panel to be dry and to have a "warmed-over" flavor. These comments by the panel members and the partial destruction of the normal red color during aging suggested that the steaks were partially cooked and that only a part of the tenderization resulted from an aging effect. Aging at 100° F was eliminated because bacterial spoilage was more difficult to control than at 110° F. The above series of experiments demonstrated that steaks could be tenderized in the manner prescribed and it was of interest to determine if the tenderization achieved was comparable to that obtained when steaks were aged for two weeks at 35° F.

High temperature aging compared to a more conventional aging procedure. Steaks were cut from 8 pairs of U. S. Good inside rounds which had been infused with oxytetracycline, 2 pairs with 20 ppm, and 6 pairs with 30 ppm. The increases in tenderness of steaks aged 24 hr at 110° F were compared to adjacent steaks aged 14 days at 35° F. The steaks were randomly assigned taking into consideration the differences between animals, sides, locations within the muscle and aging temperature.

After aging at 110° F, steaks from two of the rounds infused with 20 ppm of oxytetracycline had somewhat higher bacterial counts than those infused with 30 ppm but all were considered satisfactory for taste panel evaluation. The taste panel and shear test results are given in Table 5. Significant differences between means are indicated.

TABLE 5

Mean tenderness, residue and shear values for steaks aged at 35° and 110° F

Time and temp. of aging	No. of judgments	Tenderness		No. of cores	Shear
		Initial	Residue		(lb)
Unaged control—(35° F)	165	5.2	5.2	81	11.2
Unaged control—(110° F)	178	5.3	5.5	80	10.8
14 days—(35° F)	166	5.9	5.8	81	10.8
Unaged control	165	5.2**	5.2**	81	11.2
1 day—(110° F)	179	6.3	6.2	78	9.3
Unaged control	178	5.3***	5.5***	80	10.8**
14 days—(35° F)	166	5.9	5.8	81	10.8
1 day—(110° F)	179	6.3*	6.2*	78	9.3**

* P<0.05.
** P<0.01.
*** P<0.001.

Tenderness was increased significantly by both aging treatments. The mean tenderness scores showed an increase of 0.7 unit on the tenderness scale for aging at 35° F and 1.0 unit for aging at 110° F. Shear values were unchanged by aging at 35° F but followed the taste panel scores for steaks aged at 110° F. The results of the Warner-Bratzler shear test in this experiment were rather typical of other comparisons in which the larger taste panel differences in tenderness were reflected in significant differences in shear values and small differences were not. From the results given in Table 5 it was concluded that under the conditions used, steaks were as effectively tenderized in 24 hours at 110° F as in 14 days at 35° F.

While this study was primarily concerned with tenderness, an objectionable flavor, described by the panel members as acrid or stale, was frequently noted in the aged steaks. The undesirable flavor occurred more frequently in steaks aged at the higher temperature. It seems likely that the large surface area of the steaks provided a greater opportunity for flavor deterioration than would the relatively limited surface area of beef rounds or sides.

SUMMARY

Round steaks from U. S. Utility and U. S. Good carcasses were used to determine the optimum conditions of high temperature and time for increasing the tenderness of beef by aging. Microbial growth during aging was effectively controlled by infusion of 30–50 ppm of oxytetracycline into the rounds prior to cutting, or by 45,500 rad of gamma radiation in combination with the antibiotic treatment.

At the level used (45,500 rad), gamma irradiation did not affect the tenderness or tenderization of steaks aged at 110° F for 24 hours.

In other experiments, 2 days at 90° F and 24 hours at 110° F were found to be optimal for aging steaks to achieve effective tenderness. Steaks aged for 24 hours at 120° F showed greater increases in tenderness than those aged for 24 hours at 110° F or for 2 days at 100° F. Aging at 90° F necessitated more severe treatment to prevent excessive bacterial growth; while aging at 120° F resulted in more undesirable color and flavor changes in the steaks than found in those aged at the lower temperatures. On this basis, 24 hours at 110° F was selected as the most desirable aging time and temperature for the rapid tenderization of beef round steaks. Additional studies showed that steaks aged for 24 hours at 110° F had tenderness increases comparable to those found in steaks aged for 14 days at 35° F.

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Time-Temperature Tolerance of Frozen Foods. XX.

Boysenberries

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(Manuscript received July 31, 1959)

DETAILED RESULTS on time-temperature tolerance of major frozen fruits in retail packages have been reported *(2, 3, 4, 5)*. This is the first paper in the series which gives results on effect of temperature in larger than retail packages of frozen fruits. Since frozen boysenberries are largely prepared in IQF (individually quick frozen) form and packaged in institutional sizes for remanufacture into pies and other products, the principal effort in the experiments reported in this paper was devoted to this type of product and size of package. This paper gives results on moisture, ascorbic acid, color, and flavor changes in both IQF and sirup-packed frozen boysenberries held at temperatures from 0° to 30° F.

MATERIALS AND METHODS

The berries used were all representative commercial packs obtained from freezing plants in California and Oregon during the 1955 and 1956 seasons. There were 8 lots of IQF and 3 of sirup-packed berries, each lot consisting of 120 to 140 lb of berries. The IQF berries were frozen in field crates in air-blast freezers at −20° to −30° F, and then separated by placing the frozen blocks between rollers. The separated fruit was inspected and sorted prior to loose packaging in 20-lb cardboard cartons. Berries for the retail packs were washed, sorted, and packed in 12-oz. composite cartons in the ratio of 3 parts berries to 1 part 50% sucrose sirup. The packages were placed on wire-mesh trays in single layers and frozen in an air blast at −20° F.

All samples were transported to the Laboratory under dry-ice refrigeration and held at −20° F until placed at experimental temperatures of 0°, 10°, 20°, and 30° F. At intervals samples of both sirup-packed and IQF berries were removed from the experimental temperatures and cooled to the control temperature of −20° F.

Organoleptic evaluation. Color and flavor changes were determined by the triangle test *(1)*. Ten to 15 trained judges were used for both color and flavor measurements and each test was replicated 2 or 3 times, giving a total of 20 to 30 judgments. Six packages of sirup-packed berries and enough IQF fruit to make four 9-in pies were used for each color and flavor comparison. Samples from the experimental temperatures were judged against −20° F controls. Comparisons involving IQF berries were made on pie filling which was prepared as follows: Berries were partially thawed in a glass mixing bowl in the draft of an electric fan for 30 min. A dry blended mixture consisting of 180 g sugar, 23 g of pre-gelatinized starch, and 1.5 g of salt was added to 650 g of partially defrosted berries and the ingredients were gently stirred by hand until uniformly blended. The filling was then placed in a 9-in aluminum pie pan and baked in a gas-fired reel oven for 30 min at 400° F. The crusts were made with unbleached pastry flour using 60% lard, 24% water, and 1.7% salt (based on weight of the flour). After baking, the pies were allowed to cool at room temperature for 2 hr before removing the top crust and proportioning the baked filling for color and flavor evaluation.

Objective measurements. Oxidized ascorbic acid (DHA + DKA) was determined by procedures described in a previous publication *(6)*. For IQF berries, the determinations were made on a composite sample taken from two 20-lb boxes and, for sirupped berries, on a composite of 4 individual packages which had been thawed 2 hr at 70° F in the draft of an electric fan. Color distribution between berries and sirup was determined on thawed berries drained on an 8-mesh stainless-steel screen for 2 minutes. The transmittancy of a water extract from the drained berries and of diluted drained sirup was determined at 515 mmc. The procedure was essentially the same as that described for raspberries *(4)*.

Color of the baked pie filling prepared from IQF berries was determined on a Gardner Automatic Color-Difference Meter. The instrument was standardized with a red tile having the following values: L = 24.4, a ⟶ = 15.7, b_L ⟶ 4.3.

Moisture loss in the IQF fruit was determined by periodically weighing the boxes held at various temperatures.

RESULTS AND DISCUSSION

Table 1 shows the range of "high-quality life" *(8)* for IQF and sirupped boysenberries. Since panel members were highly trained and gave reproducible results for a given treatment, the ranges shown in Table 1 can be ascribed largely to differences in product characteristics inherent in the different lots. These differences include varying maturity level, seasonal or climatic effects, cultural and harvesting practices, processing variables, and many others. It should be emphasized that the ranges given are based on only 2 seasons and a limited number of lots. Consequently, it is possible that high-quality life of some commercial samples may be shorter or longer than indicated. Nevertheless, the data clearly show the importance of temperature, and this approximate temperature effect would apply to samples outside the ranges given as well as to those within.

TABLE 1

Effect of temperature on high-quality life of IQF and sirupped boysenberries as determined by trained taste panels [1]

Temperature	Range in days	
	Flavor	Color
IQF berries		
0	360–450	270–360
10	100–150	100–150
20	40–50	40–50
30	12–16	10–20
Sirupped berries		
0	550–750	
10	140–180	140–180
20	28–42	14–28
30	4–7	10–16

[1] High-quality life is defined as the time elapsed before a change in flavor or color is detectable by 65% of the judges.

Figure 1 indicates an essentially exponential relation between rate of flavor change and temperature.

[a] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

This function of temperature is in agreement with results obtained on peaches, strawberries, and raspberries *(2, 3, 4)*. However, siruped boysenberries are somewhat more stable than raspberries and considerably more so than strawberries and peaches.

Because of original variability in color of IQF berries and additional variables introduced during preparation for, and baking in, pies, no consistent trends attributable to time or temperature were observed in color as measured by Gardner meter readings of the baked pie filling.

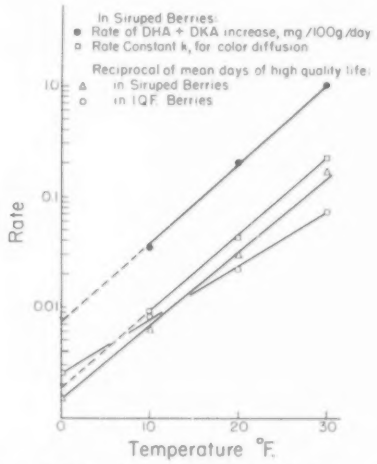

Figure 1. Relationship between rate of flavor, color, and oxidized ascorbic acid changes and temperature.

With siruped berries, however, a relationship between diffusion of color from berries to sirup and temperature history was established. The color diffusion data were plotted on semilogarithmic paper and are shown in Figure 2. The expression $(R + 1)/(R - 1.4)$ comes from an equation which was derived for the color diffusion process in raspberries *(7)*. The equilibrium color ratio of 1.4 is not a rigorous value because of the limited number of samples on which it is based. However, the straight lines obtained with this equilibrium color ratio indicate that color diffusion in boysenberries follows a first-order process just as it does in raspberries and hence affords a means of determining "equivalent temperature history." As explained in the paper on raspberries *(7)*, the rate constant and color ratio permit calculation of equivalent exposure time to some chosen steady reference temperature.

Ascorbic acid changes were expressed as increase in dehydroascorbic acid and diketogulonic acid (DHA + DKA) with time (Figure 3). As for strawberries in composite cartons *(3, 6)*, total ascorbic acid (ASA + DHA + DKA) remained constant throughout the study and ranged from 18 to 21 mg/100 g in both siruped and IQF berries. Initial DHA + DKA ranged from 2 to 3 mg/100 g in IQF berries and 5 to 6 mg/100 g in the siruped product. The time required for 10 to 12 mg/100 g of DHA + DKA to accumulate in

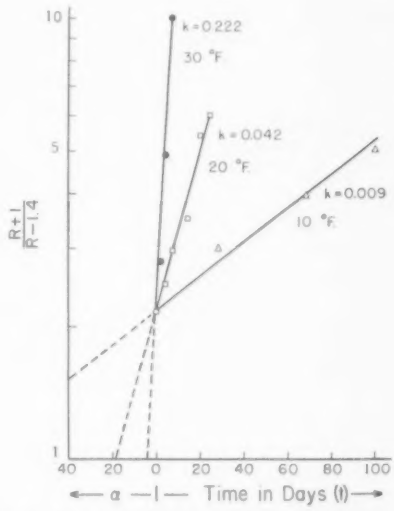

Figure 2. Color diffusion in siruped boysenberries. Relationship among the parameters in the equation $\ln \frac{R+1}{R-1.4} = k(t+\alpha)$ where R = color ratio (fruit/sirup), 1.4 = equilibrium color ratio, k = rate constant, and α = time at the various temperatures to equal prefreezing diffusion.

either siruped or IQF berries corresponded to the approximate high quality life. This observation suggested that measurement of DHA + DKA could be used as a guide for the assessment of relative flavor quality in frozen boysenberries. Rates of ascorbic acid oxidation in IQF berries were approximately twice as high as in siruped fruit at 10° to 20° F but at 30° F the difference was much smaller. These results are probably due to the fact that most of the sirup at the lower temperatures was still frozen and hence exerted its maximum effect in preventing oxidation of the berries.

For siruped berries, rate of ascorbic acid oxidation, color diffusion, and flavor change plotted against temperature on semilogarithmic paper all yield straight lines which are essentially parallel (Figure 1). Calcu-

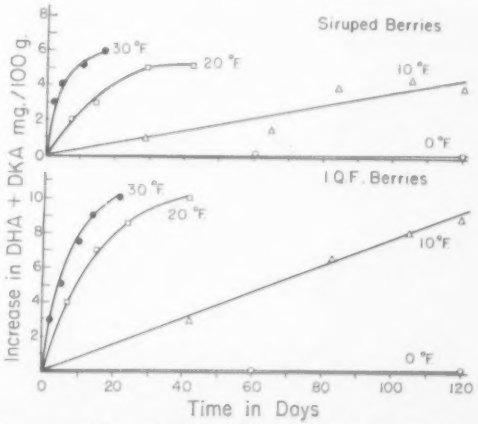

Figure 3. Effect of time and temperature on ascorbic acid oxidation in IQF and siruped boysenberries.

lated energies of activation in the 10° to 30° F range were of the order of magnitude of 41.3, 42.0, and 43.6 kcal for flavor change, color diffusion, and ascorbic acid oxidation, respectively. Thus, it appears that the temperature coefficients for all three measurements are very similar.

Moisture loss in loosely packed IQF berries in slip cover cardboard cartons is shown in Figure 4. At 0°

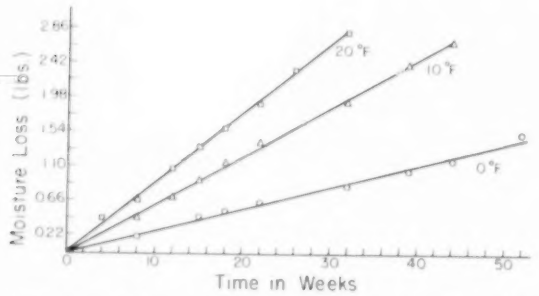

Figure 4. Effect of time and temperature on moisture loss from IQF boysenberries packed in 20-lb cartons.

to 20° F, weight loss in the 20-lb boxes was linear with time. At 30° F, the rate of moisture loss was considerably greater than at 20° F but was complicated by loss of juice due to partial thawing as well as moisture evaporation. About 1.2 to 1.6 lb of moisture, representing 6 to 8% of the package contents, was lost during high quality life of the berries at 0° to 10° F. Only 2 to 3% of the berry weight was lost as moisture during the high quality life at 20° F. Therefore, it appears that weight loss during high quality life at relatively high temperatures is minor in comparison to that which occurs at lower temperatures, because deteriorative changes in color and flavor occur much more rapidly at the higher temperature. The principal appearance changes due to desiccation were evident in the top layers. Subsurface berries appeared to be normal in size and shape. The badly desiccated berries had a more pronounced off flavor but, due to the low ratio of desiccated to normal berries, overall flavor of a given pie filling was not greatly affected. This is also indicated by the fact that approximately the same degree of flavor change was obtained in samples with moisture losses of 2 to 3 and 6 to 8%. Nevertheless, from the standpoint of actual weight loss and appearance of frozen berries, it would seem desirable to employ a more protective packaging for IQF boysenberries.

SUMMARY

Stability of sirruped boysenberries in retail packages was very good in the temperature range of 0° to +10° F, but IQF boysenberries loosely packed in cardboard cartons were somewhat less stable at the same temperatures. At 20° to 30° F the apparent stability was in favor of the IQF fruit. Color diffusion from berries to sirup provides a means of estimating temperature history in the retail packs. Time required for accumulation of 10 to 12 mg/100 g of oxidized ascorbic acid (DHA + DKA) corresponded to the approximate high-quality life (as determined by panel measurements) of both sirruped and IQF berries held at temperatures of 10° to 30° F. Moisture loss in IQF berries detracted from appearance of the top layers of fruit and reduced the package net weight by 6 to 8% during the high-quality life of samples held at 0° to 10° F. At 20° F, definite quality changes occurred when only 2 to 3% weight loss was recorded.

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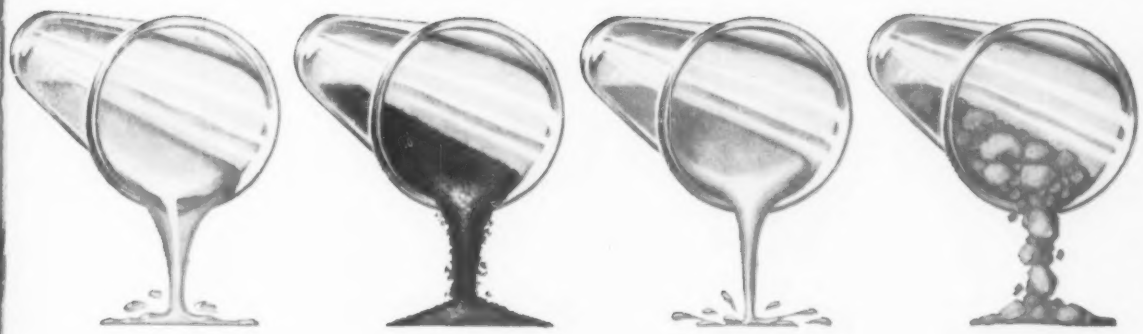

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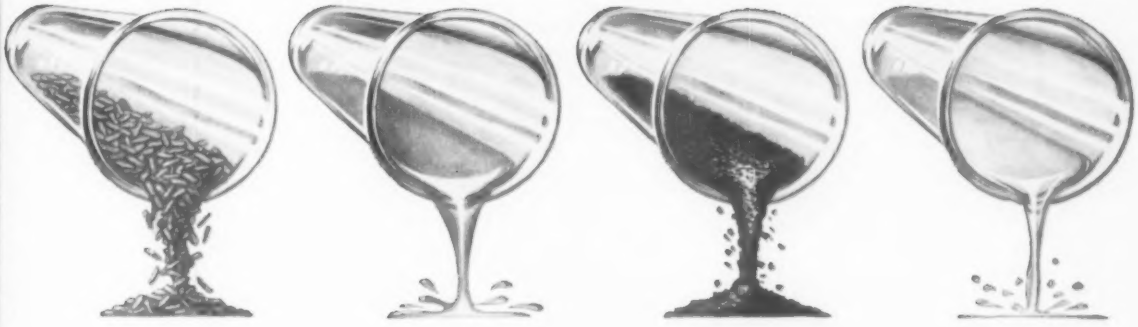

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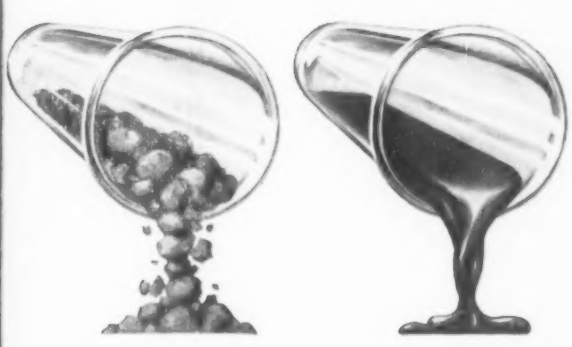

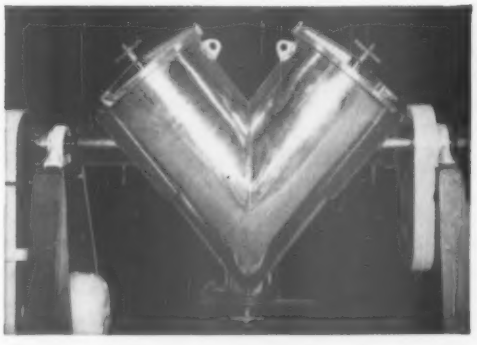

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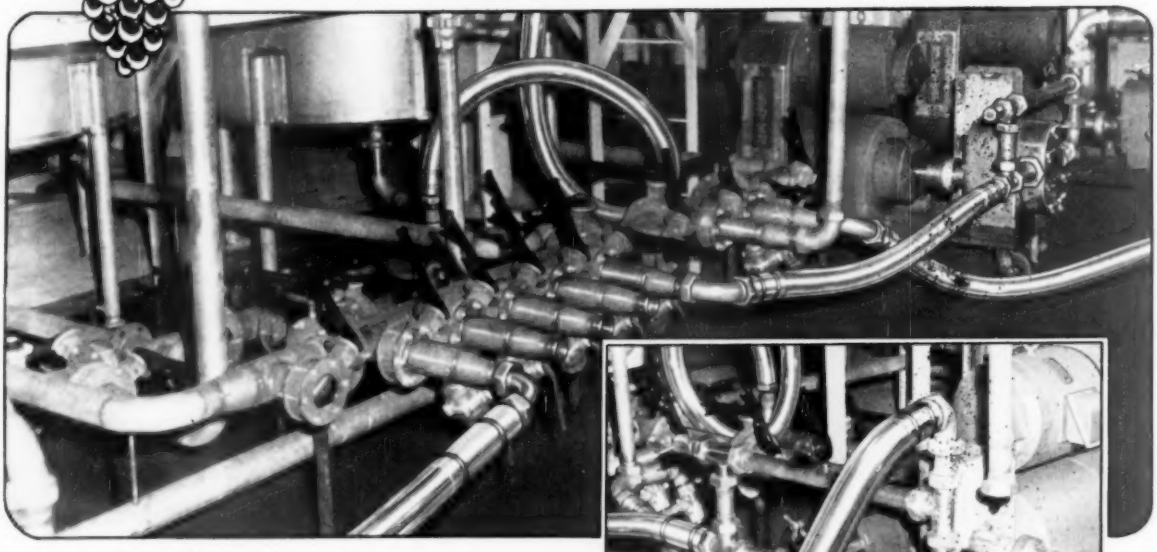

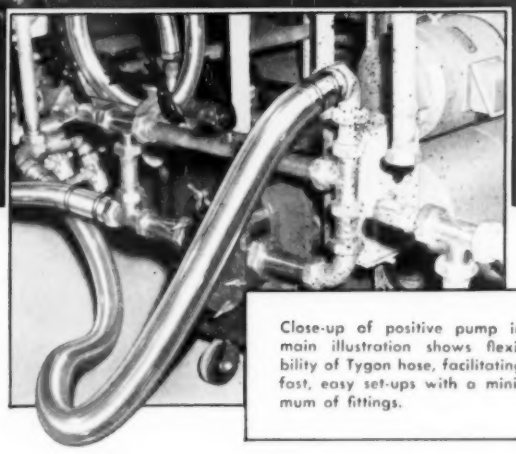

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Composition of Cherry Seed and Cherry Seed Oil *(Prunus cerasus)*

(Manuscript received September 22, 1959)

K. G. Weckel and H. D. Lee

Department of Dairy & Food Industries, University of Wisconsin, Madison

THE PROCESSING OF CHERRIES results in the disposal of very large quantities of cherry seeds. In Wisconsin and Michigan, the total cherry production averages 120 million pounds annually *(6, 13)*. The seeds are not used to provide income to the canners or growers. The kernels from cherry pits have been reported to contain an oil resembling that of almond *(15)*. The amount of oil in the kernel ranges from about 30 to 38% of the dry kernel weight *(11, 12, 17)*. The weight of the kernel represents 28% of the total cherry pit weight *(19)*. The moist cherry pit accounts for about 10% of the total cherry weight *(5)*. The weight of the moist cherry pit is about twice the weight of the dried cherry pit. From these data, it has been estimated the amount of cherry kernel oil potentially available in Wisconsin and Michigan is 100,000 gallons.

Rabak *(15)* made a study of the utilization of cherry by-products, including cherry pits from imported cherries, and from cherries grown in this country. Others have determined some of the 'fat constants' of cherry kernel oil *(1, 2, 11, 12, 14, 17)*. In the past there has been disagreement about the keeping quality of cherry kernel oil *(2, 11)*. More data are therefore needed in order to determine the usefulness and potential of cherry kernel oil and cherry kernel meal. With this in mind, the composition of cherry seed was determined and the more useful 'fat constants' of cherry kernel oil measured.

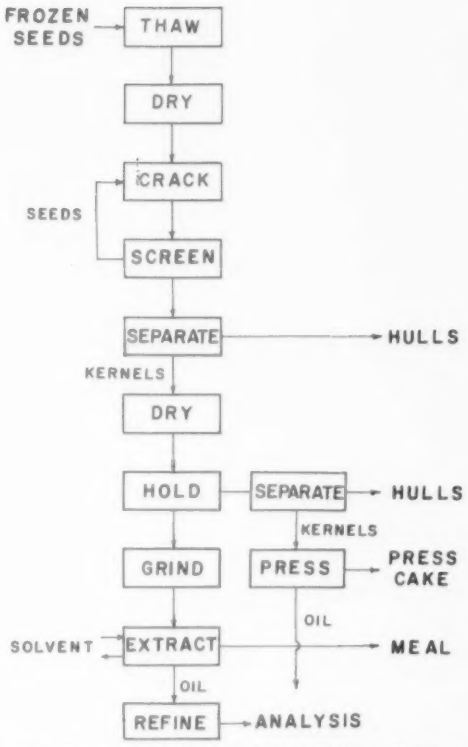

Figure 1. Preparation of cherry kernel oil from cherry seeds.

EXPERIMENTAL METHOD

1. Preparation of cherry seeds and cherry kernel oil. Cherry seeds were collected directly from the cherry pitting machines at the Fruit Growers Co-operative, Sturgeon Bay, Wisconsin. The drained seeds were frozen in burlap bags and stored at −10° F until used.

After thawing and air drying at room temperature, the cherry seeds were cracked in a Fitzpatrick comminuting machine; uncracked seeds were screened and recycled. The kernels were separated from the cracked hulls by floating them in a 20% sodium chloride solution. After washing the kernels thoroughly, they were air dried and stored at 45° F. The process is illustrated in Figure 1.

The cherry kernels were ground to about 25-mesh size and the oil extracted by use of Skelly solvent B in a Soxlet extraction apparatus. Four pounds of cherry kernels could be processed in the extractor. The crude oil was stored at −10° F until refined. The refining process for the crude oil is illustrated in Figure 2. The refined oil (refined in 100 gram quantities) was stored at −10° F under nitrogen.

Methods of analysis and results. The composition of cherry seeds given in Tables 1 and 2 was determined according to the methods of the Association of Official Agricultural Chemists. The oil content, however, was determined with a Soxlet extraction apparatus. The 'fat constants' of cherry kernel oil presented in Table 3 were determined according to the methods of the Association of Official Agricultural Chemists. The composition of the unsaturated acids present in cherry kernel oil was calculated from the percentage saturated acid, the thiocyanogen number and the iodine number. The data are presented in Table 4. The presence of oleic acid was confirmed by an elaidin test. The presence of linoleic acid was confirmed by the formation of tetrabromostearic acid from the unsaturated acid fraction. Ultraviolet spectra technique was also used to detect the presence of linoleic and linolenic acids.

Jamieson *(11)* reported that the cherry kernel is 28% of the whole seed and that the oil content in the kernel was from 32 to 40%. The results reported in Table 2 agree with these data very well. In Table 3 is presented a comparison of the composition of soybeans, almonds and cherry kernels as examined in this study.

The total nitrogen content (protein) of cherry kernels compares favorably with that of almonds and soybeans. Soybean and other meals have been used as a feed for poultry, swine and cattle *(3)*. The high protein content of cherry kernel therefore gives it potential as a feed.

The results of the analysis of cherry kernel oil are presented in Table 4. A comparison of these data is made (Table 5) with that of other investigators. The data reported by Jamie-

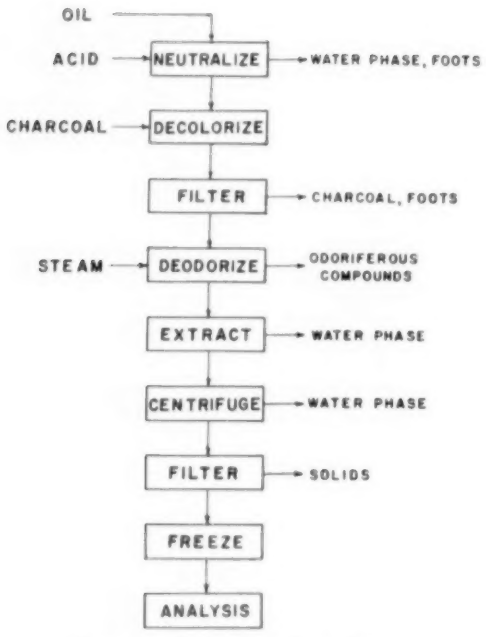

Figure 2. Refining of cherry kernel oil.

son *(11)* were based on an analysis of refined cherry kernel oil. The data reported by Stein *(18)* and by Eichenberger *(9)* were derived from cherry kernel oil prepared in this study. This oil was extracted from pits obtained the year previously and stored at 45° F. The major discrepancies in the data are in the iodine numbers, and in per cent acids. Differences in the methods used for the determination of the iodine number and in the time when the analyses were made could account for the different values reported.

In Table 6 is presented a comparison of analyses of the acids in cherry kernel oil. The linolenic acid found in this study and reported in Table 6 was only presumed to be in a quantity less than 3%. The presence of elaeostearic acid (an isomer of linolenic acid) was established as a natural constituent of cherry kernel oil. In Figure 3 is shown a typical ultraviolet spectrum of isomerized cherry kernel oil. The appearance of a peak at 234 mμ indicates diene conjugation and hence the presence of linoleic acid *(10)*. There are also peaks at 260, 270 and 280 mμ indicating the possible presence of linolenic acid as a natural constituent. In Figure 4 is shown

TABLE 1

Composition of cherry seeds

	Wet basis[1]	Dry basis[1]
Moisture	42.0	2.1
Kernels	33.4	27.0
Hulls	66.6	73.0
Crude oil	6.0	10.4
Crude protein (N x 6.25)	4.1	7.6
Ash	0.4	0.6
Starch	1.6	2.8
Sugar (invert)	0.1	0.2

[1] Per cent of the whole seed.

TABLE 2

Composition of cherry kernels and hulls

	Kernels (%)	Hulls (%)	Seeds (%)	Basis
Moisture	....	....	42.0	As received
Kernels	....	....	27.0	Moisture free
Hulls	....	....	73.0	Moisture free
Crude oil	38.4	1.0	10.4	Moisture free
Crude protein (N x 6.25)	42.7	0.7	8.5	Moisture and fat free
Ash	2.1	0.3	0.6	Moisture and fat free
Starch	17.0	....	3.1	Moisture and fat free
Sugar (invert)	1.0	....	0.2	Moisture and fat free

TABLE 3

Comparison and composition of cherry kernels, soybeans, and almonds

	Cherry kernels (%)	Soybeans[1] (%)	Almonds[2] (%)	Basis
Crude oil	38.4	19.6	56.6	Moisture free
Crude protein	42.7	53.2[4]	45.2[4]	Moisture and fat free
Ash	2.1	6.2	7.3	Moisture and fat free
Total carbohydrates	18.0	9.9[3]	47.6	Moisture and fat free
Fiber	....	6.9	6.4	Moisture and fat free

[1, 2] Altschul.
[3] Reported as sucrose.
[4] Reported as total nitrogen.

TABLE 4

Characteristics of cherry kernel oil

	Expressed	Solvent extraction
Specific gravity	0.916(25/25°)	0.916(25/25°)
Refractive index	1.4746(20° C)	1.4762 (20° C)
Free fatty acid	1.2	0.08
Acid number	2.4	0.16
Peroxide value	not detectable	2.14
Saponification number	184	187
Soluble acids	.....	0.3%
Insoluble acids (Hehner No.)	.....	94.0%
Unsaponifiable matter	.....	0.84%
Iodine number	.....	103.3
Thiocyanogen number	79.9	79.3
Saturated acids	.....	7.4%
Unsaturated acids	.....	86.4%

TABLE 5

Composition of cherry kernel oil as reported by different investigators

	This study[1]	Jamieson[2]	Eichenberger	Stein	Wells[3]
Specific gravity	0.916(25/25°)	0.918(25/25°)	.....	.....	.....
Refractive index	1.4762(20° C)	1.4740(20° C)	.....	1.4739(25° C)	.....
Acid value	0.16	0.09	.....	1.33	
Free fatty acid	0.08	.....	0.66	0.66	
Peroxide value	2.14	.....	0.82	.....	.....
Saponification number	187	190.7	187	187	.....
Neutralization equivalent	.....	.....	.....	.....	281.7
Soluble acids	0.3%	.....	.....	.....	.....
Insoluble acids	94.0	.....	.....	.....	.....
Reichert Meissl value	.....	0.3	.....	.....	.....
Polenske value	.....	0.2	.....	.....	.....
Unsaponifiable matter	0.84%	0.5%	.....	.....	1.3%[4]
Iodine number	103.3 (Wijx)	115.8 (Hanus)	92.1	108.6 (Wijs)	112.2
Thiocyanogen number	79.9	.....	.....	.....	.....
Saturated acids	7.4%	7.7%	.....	28.4%	16.4%
Unsaturated acids	86.4%	87.0%	.....	67.2%	83.6%

[1] Solvent extracted and refined. [2] Refined. [3] Values are reported on the fatty acids of the oil. [4] Oil basis.

TABLE 6

Composition of fatty acids in cherry kernel oil

	This study	Stein	Wells[1]	Gas chromatography[2]
Saturated acids	7.4%	28.4%	16.4%	10.1%
Unsaturated acids	86.4%	67.2%	83.6%	89.8%
Palmitic acid	.....	.....	.....	7.7%
Stearic acid	.....	.....	.....	2.4%
Oleic acid	60.8%	32.1%	45.6%	55.3%
Linoleic acid	30.6%	17.8%	35.5%	34.5%
Linolenic acid	3.0%	17.3%	2.5%	Trace
Arachidonic acid	.....	—0.1%	.....	.....
Elaeostearic acid	Present	.....	.....	.....

[1] Reported as % fatty acids on a fatty acid basis.
[2] Reported as % methyl ester on a methyl ester basis. Analysis made by Wilkens Instrument & Research, Inc.

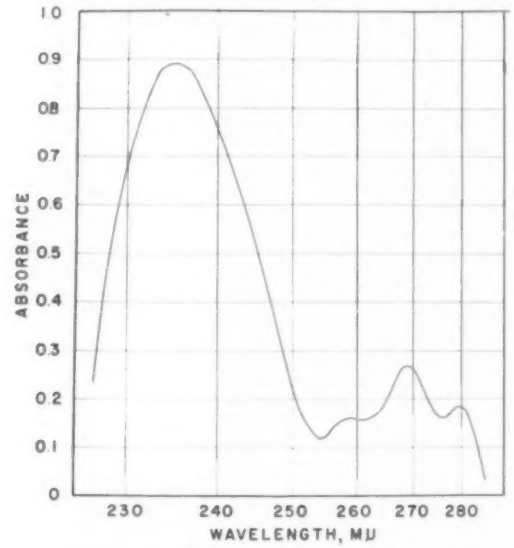

Figure 3. Ultraviolet spectrum of isomerized cherry kernel oil.

the ultraviolet spectrum of un-isomerized cherry kernel oil. The peaks at 260, 270 and 280 mme indicate the presence of elaeostearic acid. The elaeostearic acid could have resulted from the isomerization of linolenic acid during storage and by exposure to light *(16)*. The ultraviolet spectrum of oil which was extracted from cherry pit in the frozen state indicates the presence of elaeostearic acid (Figure 5). Because of the absence of peaks at 310 mme in the ultraviolet spectra, arachidonic acid was presumed to be absent. Stein reported the presence of arachidonic acid in the sample submitted to him *(18)*. Wells reported the presence of linolenic acid but made no mention of elaeostearic acid *(20)*. Gas chromatographic analysis of a sample of cherry kernel oil prepared in this study indicated presence of trace amounts of linolenic acid *(7)*.

COMPARISON WITH OTHER OILS

In Table 7 is presented a comparison of the composition of cherry kernel oil with other oils closely resembling that of cherry kernel oil. In general, cherry kernel oil is somewhat like corn oil and peanut oil.

SUMMARY

The composition of cherry seeds and of cherry kernel oil was determined.

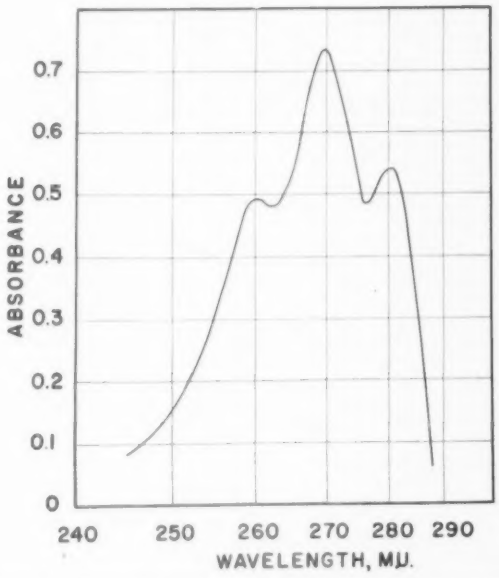

Figure 4. Ultraviolet spectrum of un-isomerized cherry kernel oil.

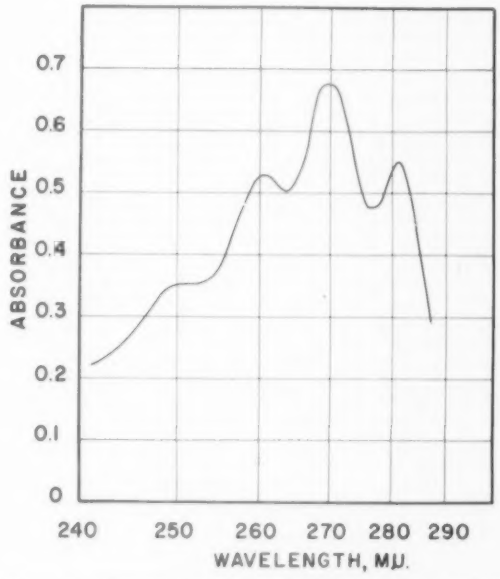

Figure 5. Ultraviolet spectrum of cherry kernel oil extracted from kernels in the frozen state.

Cherry kernels have a fairly high protein and oil content which may have potential in animal and food mixtures. The composition of cherry kernel oil resembles that of almond oil and is quite similar to that of corn oil, peanut oil and sesame seed oil.

Elaeostearic acid was shown to be present in cherry kernel oil. The presence of elaeostearic acid may be the cause of accelerated development of oxidative rancidity observed in the oil.

TABLE 7

Composition of cherry kernel oil and other oils

	Cherry kernel oil[1]	Almond oil[2]	Corn oil[2]	Peanut oil[2]	Poppyseed oil[3]	Sesame seed oil[3]	Sunflower seed oil[3]
Iodine number	103	95–102	117	83–95	132–142	104–116	139
Saponification number	187	183–196	190	185–192	188–196	187–193	190
Myristic	.....	1.2	.....	.....	.....	.....	.....
Palmitic	7.7	4.5	7.3	6.0	4.8	8.2	6.4
Stearic	2.4	.....	3.3	4.7	2.9	3.6	1.3
Arachidic	.....	.....	0.4	3.2	.....	1.1	4.0
Lignoceric	.....	.....	0.2	2.5	.....	.....	.....
Oleic	60.8	77.0	43.4	58.0	30.1	45.3	21.3
Linoleic	30.8	17.3	39.1	20.7	62.2	41.2	66.2
Linolenic	.....	.....	.....	.....	.....	.....	.....

[1] This study.
[2] Jamieson (*11*).
[3] Eckey (*8*).

Acknowledgment

The authors wish to thank Dr. K. P. Dimick of Wilkens Instrument & Research, Inc., for analyzing a sample of cherry kernel oil by gas chromatography, and to Dr. J. A. Stein, Pillsbury Mills, Inc., and W. R. Eichenberger, A. E. Staley Mfg. Co., for analyses of samples of oil submitted to them.

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Flor Sherry Production by Submerged Culture

(Manuscript received September 19, 1959)

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THE CONVENTIONAL METHOD of flor sherry production, growing a film of flor yeast on the wine surface under aerobic conditions, requires five years or more for a desirable flor character to develop. Various trials with larger surface-to-volume ratios have had such slight success that there has been little change in winery techniques.

The flor character has not been chemically analyzed, but experienced tasters recognize the odor without difficulty. Aldehyde formation during fermentation, having some correlation with the intensity of the flor character, is a criterion for chemical measure of the flor flavor—a crude system at best. A more specific method of analysis is needed.

The main pathway of production of acetaldehyde is by the action on alcohol of alcohol dehydrogenase (ADH) and coenzyme I (DPN) to give the aldehyde and reduced coenzyme I. Equations (1, 2) show the relationships.

$$\text{Ethyl alcohol} + \text{DPN} \xrightleftharpoons{[\text{ADH}]} \text{Acetaldehyde} + \text{DPNH}_2 \qquad (1)$$

$$\text{DPNH}_2 \xrightarrow{[\text{O}]} \text{DPN} + 2\text{H} \qquad (2)$$

Fornachon *(8)*, using several yeasts other than *Saccharomyces beticus*, showed that this reaction takes place in aerobic fermentations.

Crowther and Truscott *(5, 6)* recently reported that flor-like wines could be produced by pumping-over wines inoculated with flor yeast. They obtained relatively low aldehyde yields, probably because oxygen was insufficient to cause the reactions to proceed farther in the formation of aldehyde from alcohol. Their experiments were carried out at atmospheric pressure.

Modern methods of submerged culture techniques have been applied by Amerine *(3)* and Ough and Amerine *(9)* to growing flor yeast, *Saccharomyces beticus*, and rapidly producing aldehydes and flor character in wine. Recommended air pressure, pH of media, and rate of media transfer and renewal have been determined. Some preliminary taste studies show that aging in oak cooperage and the addition of some sugar are necessary to a superior taste in the finished product.

The present studies were conducted to define more clearly such conditions of production as aeration, alcoholic content of the medium, and the effects on the yeast population of initially starting an aldehyde fermentation. In addition, a critical taste study was made to compare flor sherries aged in glass for one year and in small oak cooperage for nine months and for one year.

EXPERIMENTAL

Equipment. The equipment used was discussed by Amerine *(1)* in detail. Figure 1 shows a schematic drawing on one of the fermenters and the pertinent equipment. It is a jacketed, baffled, stainless-steel tank with an operational capacity of 96 liters and a head space volume of 83 liters. Temperature can be controlled within ±0.1° C over a range of 9° C to 100° C by circulating heated or cooled water through the jacket. A variable-speed drive, atop the tank, operates either propeller or flat paddles over the respective ranges of 182–396 rpm and 170–300 rpm. The medium can be pumped-over and sprayed through the headspace by a variable-speed pump at rates of 2.8 to 4.4 gal per minute. Gas can be sparged through an open $\frac{3}{8}$-in tube with orifice below the stirring blades and between their sweep and the wall of the tank. Pressure on the tank is maintained by an air regulator that controls within ±0.5 psig over a range of 0–100 psig.

Media. The medium for the fermentations was wine made at the enology laboratory from grapes grown at the University of California vineyards at Davis. The wine used in the 1959 experiments, from a single lot of Palomino grapes, was adjusted to a lower pH with sulfuric and tartaric acids. Wines for the 1958 experiments were made from various types of grapes, without adjustment of pH.

Fermentation conditions. The conditions maintained during the 1959 experiments included 15 psig air pressure, with about 0.1 liter per minute air through-put, and either stirring 15 min per hour with flat paddles at 297 rpm or continuous pumping-over at a rate of 3.4 gal per min. In both years the tank fermentation temperature was 65° F. Conditions for all the 1958 experiments were 15 psig air pressure, 0.1 liter per min air through-put, and stirring 15 min per hr with flat paddles at 297 rpm except in one series, in which variable pressures were tested.

Yeast culture. Culture build-up of *Saccharomyces beticus* was accomplished by anaerobic fermentations of the yeast under static conditions at 75° F in sterile grape juice diluted to 10% sugar. When the fermentative action of the culture had ceased, a 5% inoculum containing 50 to 75 x 10^6 cells per ml was added to the medium.

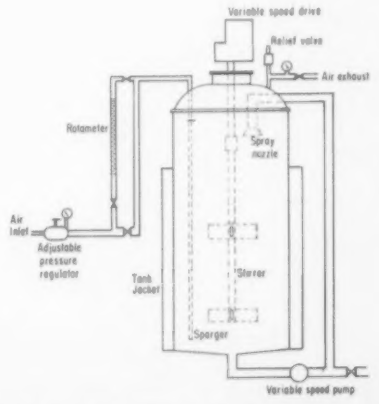

Figure 1. Schematic diagram of controlled fermentation unit.

Analyses. Chemical analyses were made by the methods of Amerine *(2)*. Standard procedures were used in determining correlation coefficents and analyses of variance, and Duncan's *(7)* multiple range test in determining significant differences in the means.

Aging and tasting. The effects on quality of aging in oak cooperage were tested by aging the flor sherries made in 1958 in 5-gal oak barrels and comparing the product with the same sherry aged in glass. In addition, samples from the barrels were drawn off at 9 months and bottled to be compared with samples aged in wood for 1 year and with samples stored in bottles for 1 year. Six experienced tasters scored these three sherries for quality on a 20-point scale. Twenty-four aged sherries were thus tasted.

Oxygen uptake studies. Rate of oxygen uptake in the fermenters was determined by the method of Cooper *et al (4)*. Briefly, 25 gal of 0.1*N* Na_2SO_3 was added to the fermentation tank with enough copper to catalyze the reaction. The method required that a sample be drawn from the fermenter, before and after each run, and excess iodine added to the sample to combine with the SO^{-2} and back-titrated with $Na_2S_2O_3$, the change in SO^{-2} concentration being proportional to the oxygen dissolved. Such measurements were made at atmospheric pressure, with stirring at relatively high air flows, and in one instance at increased pressure. The same method was used in determining the oxygen absorbed during the spraying of liquid through the headspace at various rates with no air flow and 15 psig pressure of air.

RESULTS AND DISCUSSION

Available oxygen. The ability of an aerobic fermentation tank to make oxygen available to microorganisms must be defined. Extensive studies in this field point out very clearly that many variables affect available oxygen: (a) shape of tank, (b) depth of liquid, (c) type of sparger, (d) air velocity, (e) the solubility of oxygen in the medium, (f) type of agitation, (g) physical properties of the medium, and (h) the air pressure used. These variables have been summarized and discussed by Steel *(12)*.

Determinations on the fermenter with the various pressures, paddles, and air flow rates showed (Figure 2) the flat paddles to be more efficient than the propeller paddles. Also, as reported by Phillips and Johnson *(10)*, increasing air pressure with stirring and sparging increase oxygen absorption proportionally to the first power of the partial pressure of the oxygen. Extrapolation from the data obtained show that absorption of the available oxygen will closely approach 100% if air flow rate is decreased to 0.1 liter per minute at 15 psig pressure, with stirring

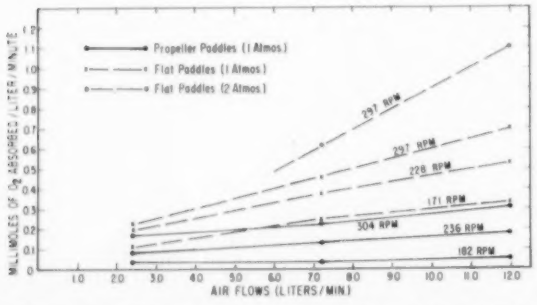

Figure 2. Rates of oxygen uptake under various paddles, paddle speeds, pressures, and gas flows for controlled fermentation equipment. Sulfite method used for evaluation.

at 297 rpm. Spraying the sulfite solution through the headspace at varying rates showed that oxygen absorption was directly proportional to the amount of solution sprayed per unit time and, in the range covered, approximately proportional to the first power of the spray rate (Figure 3). Though spraying is not as effective as stirring, fermenters using this spraying technique for fermentations with low oxygen demands are cheaper to construct and to operate.

Solomons and Perkins *(11)* showed that the efficiency of oxygen absorption declines as the use of available oxygen approaches 100%. Though this fact is generally important in aerobic fermentations with high oxygen demands, through-put flows of air in this work had to be kept low to avoid the removal of desirable volatile constituents.

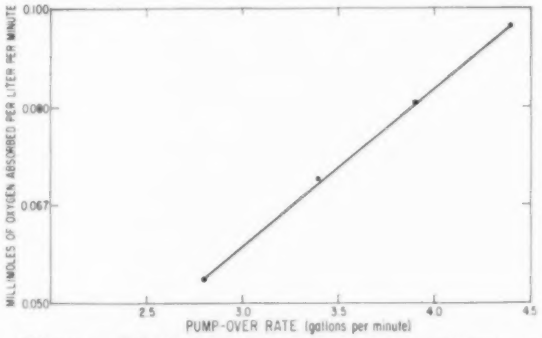

Figure 3. Relationship of pump-over rate to millimoles of oxygen absorbed under 15 psig air pressure and pump-over liquid sprayed through headspace. Sulfite method used for evaluation.

Flor sherry fermentations. So that the alcohol content of the medium could be more clearly related to the resulting aldehyde concentration produced by the fermentation, ten experiments were carried out at various alcohol levels. The fermentations, all with the same medium and at the same conditions, were fermented until aldehyde concentration reached a maximum and remained constant for several days. In the range covered, the alcohol concentration of the medium showed a correlation coefficient of −0.788 to the aldehyde concentration of the final product, significant at the 1% level (Figure 4). In previous work *(9)* an alcohol concentration below 15.0% by volume has been recommended. The soundness of this recommendation is substantiated by these results.

A second experiment was conducted to determine if pumping-over was as satisfactory as stirring as a method of aeration. Differences in rates of aldehyde production were slight (Figure 5). Yeast cell counts made during the two fermentations (Table 1) indicate a much greater production of yeast cells in the stirred than in the pumped-over fermentation. More yeast cells were available for aldehyde formation in the stirred sample, but there was no significant difference in aldehyde production between the two methods of aeration. These data indicate that pumping-over is a satisfactory method of aeration for this process.

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TABLE 1

Effects of stirring versus pumping-over on yeast cell count in aldehyde fermentation

Days	Total yeast count[1] × 10^6 cells/ml		Viable yeast count[2] × 10^6 cells/ml		Viable budding cells Per cent		Dead cells Per cent	
	Stirred[3]	Pumped-over[4]	Stirred	Pumped-over	Stirred	Pumped-over	Stirred	Pumped-over
Start								
0	.....	.....	.....	.....	....	....	....	....
1	0.35	0.20	0.02	0.05	29	25	43	75
4	0.65	0.15	0.35	0.10	70	50	46	33
7	0.55	0.95	0.15	0.45	67	56	73	53
11	1.80	0.80	1.60	0.45	44	55	13	44
14	3.00	0.70	2.05	0.40	71	75	31	43
18	3.40	0.90	0.90	0.45	67	67	74	50
21	5.45	0.25	2.55	0.25	76	60	53	....
28	5.34	.....	0.50	.....	0	0	91	100

[1] Total yeast count obtained by dilution and counting in hemocytometer.
[2] Viable yeast count obtained by staining with methylene blue and counting in hemocytometer.
[3] Stirred 15 min/hr under 15 psig air pressure.
[4] Pumped-over continuously under 15 psig air pressure.

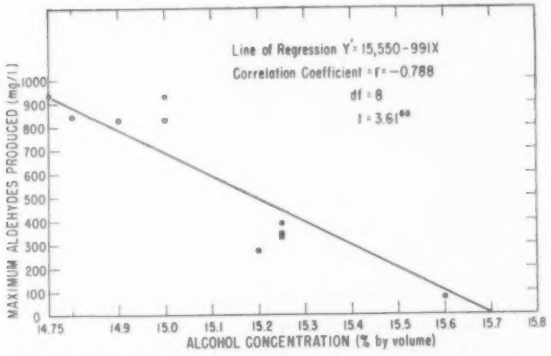
Line of Regression Y' = 15,550 - 991X
Correlation Coefficient = r = −0.788
df = 8
t = 3.61**
MAXIMUM ALDEHYDES PRODUCED (mg/l.)
ALCOHOL CONCENTRATION (% by volume)

Figure 4. Relationship of alcohol concentration in the medium to the maximum aldehyde concentration produced. Correlation coefficient calculated by method of least squares. Calculation by "Student's t" shows correlation coefficient to be highly significant.

They further indicate that the amount of viable yeast cells present at these concentrations is not a limiting factor for aldehyde formation.

Several methods are available for starting these aerobic fermentations: (1) add an active film culture to dry wine; (2) start fermentation with grape juice and, after fermentation is complete, allow yeast to go into the aerobic state, and (3) add an active anaerobic culture to dry wine. Methods 2 and 3 are essentially the same since both require time for the yeasts to acclimatize themselves to the aerobic state. Method 1 gives faster results because the yeast in the film stage is growing aerobically. Actually, if the methods are all started from a slant at the same time, they should reach the aerobic stage in the final medium at the same time. Method 1 has the advantage of not tying up the fermentation tank for a long initial anaerobic fermentation and acclimatization period.

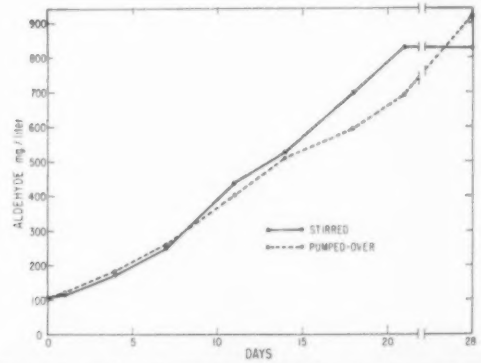

Figure 5. Rates of aldehyde production by two methods of aeration—pumping-over, and stirring. 15 psig air pressure. Stirred 15 min/hr at 297 RPM. Pumped-over at 3.4 gallons/min continuously. Temperature maintained at 65° F.

To determine whether the initial slow change from anaerobic to aerobic fermentation was truly acclimatization or the development of a mutant strain of yeast present in small amounts, viable yeast cell count was followed during the change in one of the fermentations freshly inoculated as described by method 3. The results (Table 2) indicate that the change is acclimatization. The viable yeast cell count remained reasonably constant after the dilution change expected on addition of the inoculum to the medium. If the viable cell count had shown a very large decrease and then increased at a logarithmic rate, the mutant yeast theory would have been the more acceptable.

Effects of aging in oak cooperage versus glass. Most wine chemists and wine makers know that aging flor sherry in wood helps the product, though the reasons for this increase in quality are not fully understood.

These experiments do not attempt to explain why aging in oak cooperage improves the flavor, but they

TABLE 2

Total and viable yeast cell counts on aldehyde fermentation started by addition of an anaerobic culture to dry wine

	Days	Aldehydes mg/liter	Total yeast cell × 10^6	Viable yeast cell[1] × 10^6
Culture[2]	....	....	59.5	46.5
Culture[3] and wine	0	20	....	0.94
	1	26	3.3	0.67
	2	32	4.1	0.84
	4	54	7.3	....
	5	....	....	0.55
	8	99	3.3	0.53
	12	198	6.0	0.61

[1] Viable yeast cell count made by dilution and plating.
[2] Culture media was sterile grape juice; yeast was *Saccharomyces beticus* grown in incubator at 25° C under static conditions.
[3] Ratio of culture to wine was 1 to 20 gallons.

definitely show that it does so. They also contribute some comparative data that may aid in future explanation of the causes of this improvement in quality.

Table 3a shows analysis of variance for the tastings conducted. Significance was found between wines, tasters, and treatments, with no significant interactions. To establish significant differences between the various means of wines, tasters, and treatments, Duncan's multiple range test was applied. Of most interest is the effect of the different storage treatments on the equality of the wines; this indicates (Table 3b) that storage in oak cooperage for 9 months or 1 year gives a higher quality than that of the same wine stored in glass for the same period.

TABLE 3a

Flor sherry aging experiment—analyses of variance

Source of variation	df	ss	ms	F		
				Calculated	Tabular 5%	Tabular 1%
Total	431	2228				
Tasters	5	140	28.00	7.41**	2.45	3.51
Wines	23	505	21.96	5.81**	1.79	2.29
Treatments	2	192	96.00	25.40**	3.23	5.18
Interactions						
Tasters × wines	115	320	2.78	1.36	1.51	1.80
Tasters × treatments	10	61	6.10	1.61	2.18	2.99
Wines × treatments	46	140	3.04	1.24	1.51	1.80
Error	230	870	3.78			

Table 4 shows some of the pertinent analytical and tasting data gathered from these experiments. Of note are the increases in extract and decreases in pH. Volatile acidity increased in the oak-aged samples, but it is questionable whether this factor increased quality. The aldehydes, both total and free, decreased in general in the oak-aged samples. The ratios of total to free aldehydes, however, could be important. These ratios were found to range from 1.58 to 1.04, with a mean of 1.22, for both the glass- and the oak-storage experiments. Comparison of the pairs gave a mean difference of 0.08 and a standard deviation of 0.063 for the mean difference, showing relatively small variations in the ratios. There is no apparent correlation between the difference of the mean scores and the differences in extract or pH with oak storage and glass storage for the twenty-four tests. It is of note, though, that the increase in total acid, decrease in pH, and increase in extract occurred fairly consistently with aging in oak. The individual mean scores for the treatment of each wine (6 tastings) included in Table 4 are not significantly different in most cases; however, it should be remembered that the mean scores for the treatments of all the wines (144 tastings) do show significant differences between the mean scores for the glass-aged and the oak-aged wines, as has been shown.

SUMMARY

The fermentation equipment used was tested by the sulfite method to show its capabilities for maintaining aerobic conditions with two types of paddles, by pumping-over with spraying through the headspace, and with increased pressure. The data show that with flat paddles nearly 100% of the oxygen would be available to the yeast with an input of 0.1 liter air per minute under the fermentative conditions described.

Data on the alcohol content of aldehyde fermentations show that alcohol content should be kept below

TABLE 3b

Multiple range test:[1] comparisons of means

Treatments	Glass (1 year)	Barrel (1 year)	Barrel (9 months)									
Mean score	12.92	14.25	14.40									
1% and 5% level												
Tasters	1	2	3	4	5	6						
Mean score	12.86	13.42	13.93	14.00	14.44	14.49						
5% level												
1% level												
Wines	24	10	21	2	9	23	20	12	6	14	3	7
Mean score	10.90	12.94	13.11	13.16	13.22	13.34	13.50	13.72	13.83	13.83	13.88	13.95
5% level												
1% level												
Wines (cont.)	11	4	8	16	18	22	19	5	1	15	13	17
Mean score	13.95	14.05	14.11	14.11	14.34	14.48	14.55	14.60	14.65	14.77	14.87	15.05
5% level												
1% level												

[1] Duncans (7) multiple range test for significant differences in the means. Means underscored by the same line have no significant difference at the level indicated. Means are arranged in order of their increasing values.

TABLE 4

Comparative analyses and mean taste scores of flor sherries aged in glass and in oak for one year

Mean taste score[1]		Total aldehyde mg/liter		Free aldehyde mg/liter		Alcohol % by volume		Extract g/100 ml		Volatile acid g.HA_c/100 ml		pH		Total acid g.HT_a/100 ml	
Glass[2]	Wood[3]	Glass	Wood	Glass	Wood	Glass	Wood	Glass	Wood	Glass	Wood	Glass	Wood	Glass	Wood
11.33	13.83	141	174	111	158	17.4	16.3	1.3	1.4	0.03	0.04	3.70	3.50	0.25	0.30
11.17	13.50	342	312	256	272	16.8	16.8	1.7	1.8	0.03	0.04	3.70	3.65	0.22	0.28
12.33	14.67	277	257	196	221	16.8	15.1	1.4	1.5	0.04	0.07	3.85	3.80	0.30	0.36
12.33	14.33	236	236	126	194	17.3	16.5	1.3	1.4	0.05	0.12	3.70	3.15	0.22	0.32
13.17	16.33	334	360	230	228	17.0	16.8	1.3	1.4	0.05	0.07	3.75	3.70	0.31	0.37
13.17	14.17	749	675	612	507	17.7	16.7	1.2	1.4	0.05	0.06	3.70	3.25	0.32	0.43
13.33	15.00	455	411	415	371	17.6	18.5	1.3	1.7	0.06	0.09	3.70	3.10	0.34	0.49
13.50	14.50	895	877	745	755	17.0	16.9	1.7	1.7	0.05	0.07	3.45	3.45	0.42	0.47
14.50	14.67	650	517	555	433	16.1	17.3	2.1	2.3	0.05	0.06	3.70	3.45	0.54	0.62
11.83	14.33	720	711	620	632	17.1	17.5	2.2	2.3	0.04	0.06	3.80	3.75	0.54	0.56
13.33	14.17	636	610	551	520	17.2	17.3	1.9	2.1	0.04	0.06	3.60	3.75	0.47	0.55
13.33	14.33	815	751	735	679	17.3	16.3	1.7	2.2	0.05	0.06	3.65	3.45	0.42	0.61
13.00	14.67	632	542	555	505	17.4	16.5	1.9	2.2	0.06	0.07	3.85	3.50	0.42	0.50
12.33	14.50	666	636	606	560	17.4	17.1	1.9	1.9	0.06	0.12	3.90	3.50	0.41	0.58
13.50	14.00	791	738	708	654	17.4	17.1	1.7	1.7	0.06	0.07	3.80	3.60	0.39	0.43
12.83	15.00	636	585	577	526	17.7	17.2	1.5	1.5	0.06	0.07	3.85	3.70	0.37	0.42
14.50	15.67	971	900	764	714	17.0	16.8	1.0	1.2	0.04	0.05	3.10	3.00	0.53	0.57
13.33	14.83	678	628	561	471	17.5	16.7	2.0	2.2	0.03	0.07	3.25	2.80	0.49	0.63
13.67	14.67	643	572	537	423	16.0	16.7	1.9	2.3	0.03	0.03	3.15	2.90	0.54	0.63
12.67	14.50	595	595	475	489	17.0	15.4	1.9	2.3	0.03	0.04	3.10	3.05	0.56	0.60
12.67	13.33	497	348	388	286	17.3	16.4	1.8	1.9	0.03	0.05	3.20	3.20	0.55	0.55
13.17	15.17	649	568	490	423	16.8	16.4	1.7	2.5	0.03	0.05	3.20	2.90	0.56	0.66
12.50	12.83	691	581	541	427	17.5	15.9	2.0	2.4	0.03	0.06	3.15	2.65	0.56	0.70
12.00	10.17	910	770	700	528	16.3	16.1	1.9	2.3	0.03	0.04	3.15	2.90	0.57	0.65

[1] Average of six experienced tasters, on a 1-20 point scale.
[2] Stored in ½-gal bottles with cap finish.
[3] Stored in 5-gal oak barrels, filled at 2 month intervals.

15.0% by volume. Correlation between alcohol content and the maximum aldehydes produced was −0.788, highly significant for the range studied.

Comparison of stirred versus pumped-over aldehyde fermentations show that the rate of aldehyde formation is about equal for either method, despite the fewer yeast cells in the pumped-over experiment.

Investigation of the delay in starting an initial aerobic aldehyde fermentation indicates that the cause is a necessary acclimatization period, not establishment of a mutant yeast strain.

Aging in oak cooperage (9 months or 1 year) produced flor sherries judged of a better quality than the same wines aged in glass. Data show some of the chemical differences between wines stored in oak and glass containers.

Acknowledgments

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Chemical Inactivation of Enzymes in Vegetables Before Dehydration[a]

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VEGETABLE DEHYDRATION generally includes scalding or blanching of material by steam or boiling water *(2, 19)* prior to dehydration. Heating in the moist state inactivates most enzymes, but it also results in loss of crispness and formation of a cooked flavor. Experience has shown that blanched vegetables are more likely to retain good flavor and color after dehydration and storage than partially blanched or unblanched vegetables *(3, 6, 19)*. Sulfite treatment, widely used in dehydration of vegetables and fruits, prevents browning and inactivates such enzymes as polyphenol oxidase *(9)* but imparts a foreign flavor to the food.

A number of vegetables are processed without blanching: dehydrated onions, garlic, and parsley *(18, 19)* and frozen peppers and pimentos *(13)*. It is possible that the strong natural flavors of these vegetables mask off-flavors developed in storage. Otherwise one must assume a fundamental difference between vegetables that require blanching and those that do not. Such an assumption does not appear justified in our present state of ignorance regarding causes and nature of off-flavors. It is probable that these differences are quantitative rather than qualitative.

Inadequate blanching can be detected by testing for residual activity of peroxidase, catalase *(3, 6, 19)* or other enzymes *(9)*. With the exception of the probable role of lipoxidase and lipase in forming off-flavors in unblanched frozen peas *(17, 20, 21)*, little is known about the reactions which cause flavor and color deterioration.

Enzyme inactivation by chemical treatment at room temperature has been explored at this laboratory as a possible means of obtaining dehydrated vegetables of high quality. The initial experiments encouraged further investigations *(7)*. The present paper describes methods developed and used for chemical enzyme inactivation and its testing.[c]

The tissues were infiltrated under a vacuum with various chemicals and the degree of inactivation of catalase, peroxidase, acetyl esterase, phosphatase, and polyphenol oxidase was determined. The primary purpose of the experiments was to determine the efficacy of these chemicals; no effort was made to minimize leaching of soluble materials from the vegetables during infiltration and washing. The influence of several acids, ethanol, and a number of surfactants alone and in various combinations at different concentration levels was studied. Nearly complete inactivation of tested enzymes was achieved in cabbage, celery, carrots, and apples. Storage stability and organoleptic evaluation of cabbage will be described in a subsequent paper *(8)*.

MATERIALS AND METHODS

The vegetables were trimmed, washed, and cut into slices or shreds about 0.1-inch thick. Solutions failed to penetrate thicker slices uniformly. Procedures were standardized for all plant materials as described below.

Infiltration solutions. Enzyme-inactivating solutions containing acid, 48% ethanol, and a surfactant were tested. The chemicals were tried singly and in various mixtures. Appropriate controls were used for each experiment including infiltration with distilled water and each individual component of each mixture. Four acids and a number of surfactants were studied. Surfactants were tried at 2 or more concentrations in the acid-alcohol mixture, and also alone in water. The effectiveness of several acids as enzyme inactivators was compared at the same normality. The effect of differencees in pH of the acid-ethanol-surfactant mixture was also tested.

Infiltration procedure. Vegetables were infiltrated within a vacuum desiccator with 4 times their weight of the test solution. Infiltration under vacuum was necessary to insure adequate penetration. This was accomplished by 3 cycles of evacuation and release of vacuum for a total of 12 min. After decanting, the plant material was rinsed with water, then infiltrated with 0.2*M* acetate buffer (pH 6.2) followed by water, each for a duration of 5 min and 2 cycles. The buffer adjusted the pH (lowered by the acid treatment) and the water flushed out the residual chemicals. Loss of soluble components resulted from this treatment.

Enzyme tests. Qualitative and semiquantitative tests for activity of catalase, peroxidase, acetyl esterase, phosphatase, and polyphenol oxidase were performed on filtered extracts from blended vegetables. Fresh, infiltrated, or rehydrated plant materials were blended for 3 min with an equal weight of acetate or phosphate buffer near pH 6. The dehydrated vegetables were blended for 6 min with a larger volume of buffer plus water adjusted to give roughly the same content of solids as the fresh material. A portion of each blended material was boiled to serve as a control; volumes were adjusted after cooling and before filtration. Cabbage extracts were boiled 7 min to insure inactivation of peroxidase. Boiling time for other materials was not critical. No significant changes in pH occurred on boiling and filtration. Reagent controls were also included in each experiment.

The enzyme activity of each material was estimated in terms of the time required for manifestation of a characteristic reaction. Activity was considered negative when no reaction was observed for 30 min or longer. The tests were done at room temperature (23° C). These rough estimates of residual enzyme activity were considered adequate for the purposes of this study.

Catalase formed oxygen in active extracts on addition of 0.5% hydrogen peroxide, but not in the boiled extracts. The

[a] This paper reports research undertaken in cooperation with the Quartermaster Food and Container Institute for the Armed Forces, QM Research and Engineering Command, U. S. Army, and has been assigned No. 1009 in the series of papers approved for publication. The views or conclusions contained in this report are those of the author. They are not to be construed as necessarily reflecting the views or endorsement of the Department of Defense.

[b] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

[c] Those who may be interested in use of these chemicals in foods should consult the U. S. Food and Drug Administration.

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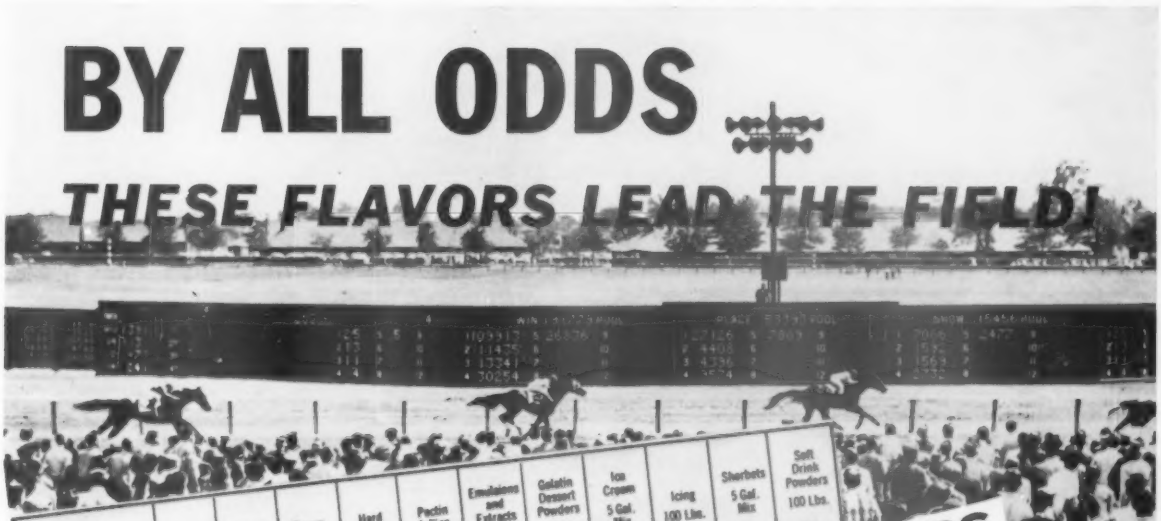

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Apricot	16.25	¼–½				4	½	½	¼	½	4
Arrack	16.00	½						½	¼	½	4
Banana	13.50	¼–½						½	¼	½	4
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time required for appearance of definite oxygen bubbles was used as a rough measure of catalase activity. Appearance of a ring of foam, if any, was also recorded.

Peroxidase activity was estimated by the method of Morris *(11, 12)*. The appearance of a blue color on the test papers treated with extract was timed. Thoroughly boiled plant extracts gave uniformly negative results.

Acetyl esterase activity was estimated with chlorophenol red diacetate as substrate at pH 6 by timing the appearance of a pink color. The substrate was prepared by acetylation of chlorophenol red (3′, 3″-dichlorophenol sulfonephthalein) *(4, 14)*. After treatment with carbon black, the chlorophenol red diacetate was recrystallized from hot methanol until free of chlorophenol red dye. Different preparations hydrolyzed enzymatically at somewhat different rates. This suggests the presence of varying amounts of chlorophenol red monoacetate. Extracts of boiled plant macerates gave no color with the substrate.

Phosphatase activity was estimated with sodium phenolphthalein phosphate as the substrate at pH near 6. Aliquots were tested at time intervals by adding an equal volume of 0.5*M* tris buffer [(tris-hydroxymethyl) amino methane], pH 8.6. Phenolphthalein phosphate hydrolyzes nonenzymatically at higher pH. A pink color was given at pH 8.5 by free phenolphthalein released in the enzymatic hydrolysis. Purification of substrate was required when reagent controls also gave a pink color when made alkaline *(5)*. The substrate was kept dry and cold and the reagent was made up fresh daily at a concentration of 0.01*M* in ethanol and acetone. Digestion time required for appearance of pink color served as a crude measure of phosphatase activity. Extracts of boiled plant macerates gave no color with the substrate.

Polyphenol oxidase activity was measurable in lettuce, apples, carrots, and celery, but was extremely weak in fresh or dehydrated cabbage. Either catechol or dihydroxyphenyl alanine (DOPA) was used as substrate. Appearance of a red-brown color was timed in a crude estimate of the activity. Extracts of boiled macerates gave no color with the substrate.

RESULTS AND DISCUSSION

Treatment of cabbage with a mixture of hydrochloric acid, ethanol, and surfactant resulted in sufficient inactivation of peroxidase, catalase, acetyl esterase, and phosphatase to give no observable reaction after 30 min (Table 1). Treatment with any one or a mixture of any 2 components did not lower enzyme activity enough to give negative or even uniformly weak tests. This shows that inactivation of the enzymes, estimated by methods used in this study, required all 3 components of the mixture.

TABLE 1

Effect of various reagents on inactivation of several enzymes in cabbage

Infiltration solutions[2]	Time required for positive reaction[1]				
	Peroxidase, seconds	Catalase		Acetyl esterase, minutes	Phosphatase, minutes
		Seconds	Foam[3]		
None (fresh cabbage)	2	6	yes	1	5
H_2O alone	2	4	yes	2	4
EtOH alone	2	5	yes	2	4
HCl alone	3	4	yes	3	5
Surfactant alone	2	4	yes	2	4
EtOH + surfactant	6	15	yes	8	15
EtOH + HCl	9	60	no	15	20
HCl + surfactant	3	42	no	12	10
HCl + EtOH + surfactant	>30 min	>30 min	no	>30	>30

[1] Negative results with boiled extracts, used as controls for each experiment, were omitted from this table. All time intervals are averages of several determinations. >30 indicates no reaction after 30 min, negative.

[2] EtOH = ethanol, 47.5%; surfactant = sodium alkyl aryl sulfonate, 0.002*M*; the same commercial lot was used in all experiments. HCl = 0.1*N* hydrochloric acid.

[3] Foam ring; yes if present, no if absent. No foam ring indicates very weak, if any, catalase activity.

Enzymes in solution or in tissue homogenates are readily inactivated by acid alone. Failure to inactivate enzymes in submerged or in vacuum-infiltrated slices with the same acid solution indicates poor penetration of the acid into living cells. This apparent lack of contact between infiltrated acid (pH 1.0 to 1.5) and enzyme proteins was confirmed by failure of tissue pH to drop below pH 3 in macerates of acid-treated and washed cabbage shreds. Nor were pieces treated with acid alone as translucent as those treated with acid-ethanol and acid-ethanol surfactant mixtures. In the latter treatment, tissue pH was 1.5 to 2.0 and enzyme inactivation was adequate by our standards. Studies on the time required for infiltration showed poor enzyme inactivation after 15 min of vacuum infiltration with acid alone, while nearly complete enzyme inactivation was achieved by infiltration with the acid-ethanol-surfactant mixture for only 1 to 2 min. Thus, we conclude that addition of ethanol and surfactant to the acid improved penetration of acid into the plant cells.

TABLE 2

Effect of infiltration solutions on time required for positive enzyme reactions in several plant tissues

Material	Infiltration solution[2]	Time required for positive reaction[1]					
		Polyphenol oxidase, minutes	Catalase		Peroxidase	Acetyl esterase, minutes	Phosphatase, minutes
			Time	Foam ring			
Cabbage shreds	H_2O + surfactant	>30	2 sec	yes	1 sec	1	10
Cabbage shreds	HCl + EtOH + surfactant (before dehydration)	>40	>40 min	no	10 min	>45	>30
Cabbage shreds	HCl + EtOH + surfactant (after dehydration)	>30	>35 min	no	>35 min	>40	>30
Carrot pieces	H_2O + surfactant	10	8 sec	yes	4 sec	5	15
Carrot pieces	HCl + EtOH + surfactant	>30	>30 min	no	>30 min	>30	>30
Celery slices	H_2O + surfactant	10	10 sec	yes	1 sec	2	>30[3]
Celery slices	HCl + EtOH + surfactant	>30	>30 min	no	>30 min	>30	>30
Apple slices (2 varieties)	H_2O + surfactant	10	1 min	yes	>40 sec	weak	>30[3]
Apple slices (2 varieties)	HCl + EtOH + surfactant	>30	>30 min	no	>30 min	>30	>30

[1] Averages of several determinations. Negative results with boiled extracts, used as controls in each determination, were omitted from this table. >30 indicates no reaction after 30 min, negative.

[2] Sodium alkyl aryl sulfonate surfactant, 0.002 to 0.005*M*. H_2O, HCl, and EtOH are distilled water, 0.1*N* hydrochloric acid, and 47.5% ethanol, respectively.

[3] No measurable phosphatase activity was found in apple and celery by the method used.

In all tested plant materials, infiltration with acid-ethanol-surfactant mixture resulted in substantial inactivation of enzymes (Table 2). Fresh, untreated celery and apple had little phosphatase activity (pH 6), and fresh cabbage appeared to lack polyphenol oxidase activity. Cabbage shreds infiltrated with acid-ethanol-surfactant mixture sometimes gave a weak but positive test for peroxidase activity. After dehydration of the cabbage, tests for peroxidase were consistently negative. Only after prolonged storage of the dehydrated cabbage, did the positive test for peroxidase return *(8)*. In cabbage at least, the enzyme peroxidase appears to be the most difficult to inactivate both chemically and by heat *(16)*.

The degree of enzyme inactivation in cabbage varied with type of acid (Table 3). As might be expected, the strong acids were more effective than those weakly ionized. Both hydrochloric and sulfuric acids were effective inactivators of cabbage enzymes, but phosphoric and acetic acids were relatively poor. The initial pH values reflect differences in the dissociation of these acids in the water-ethanol-surfactant mixture.

More detailed results on the effect of pH with hydrochloric acid are summarized in Table 4. As anticipated, the higher the pH of the hydrochloric acid-ethanol-surfactant mixture, the poorer was the enzyme inactivation in the cabbage shreds, confirming earlier observations that solutions with a pH above 2.0 allowed the infiltrated cabbage to retain considerable enzyme activity. When the pH was 3 or higher, inadequate inactivation of enzymes was the result. The effect of pH on inactivation of enzymes in cabbage agreed with information in the literature and with well-known biochemical experience.

A number of anionic, cationic, and non-ionic surface-active compounds were tried in the acid-ethanol mixture for inhibiting enzymes in cabbage shreds. Many of these chemicals enhanced the enzyme inhibition considerably (Table 5). The most effective substances were some of the anionic surface-active agents, especially the alkyl aryl sulfonates and a sodium sulfate derivative of 2-ethyl hexanol-1. The latter, however, was impure and imparted an unpleasant odor and a brown color to the treated cabbage. The compounds tested are listed in Table 5 approximately in order of their effectiveness within the classification groups. Chemical names were obtained from the literature *(10)* or deduced from the chemical composition given by the manufacturers. To conserve space, results in Table 5 were presented for one concentration only, although the majority were tested at two or more levels.

In most cases, the enzyme-inactivating effect of surfactant increased with higher concentration. When tested alone in water solutions at the concentrations used in the mixture, these surfactants appeared to have only a negligible effect on enzyme activity. The reported denaturing effect of many surface-active agents on proteins *(1, 15)* was not

TABLE 3

Effect of different acids in infiltration mixture on enzyme activity of cabbage macerates (pH 5.2)

Infiltration solution			Time required for positive reaction[1]				
Components of mixture[2]	Acid conc. *N*	Initial pH	Peroxidase seconds	Catalase		Acetyl esterase minutes	Phosphatase minutes
				Seconds	Foam ring		
H_2O alone	0.0	7.0	2	2	yes	1	4
Surfactant I in H_2O	0.0	6.4	2	7	yes	3	4
H_3PO_4 + EtOH + surfactant I	0.1	2.4	6	60	no	15	13
CH_3COOH + EtOH + surfactant I	0.1	3.1	4	21	no	4	3
CH_3COOH + HCl + surfactant I	0.1	1.6	4	20	no	8	15
HCl + EtOH + surfactant I	0.1	1.3	5 min	180	no	>30	>30
H_2O alone	0.0	7.0	2	10	yes	2	3
Surfactant II + H_2O	0.0	5.8	5	30	yes	2	3
HCl + EtOH + surfactant II	0.1	1.3	120	180	no	>30	>30
H_2SO_4 + EtOH + surfactant II	0.1	1.2	>30 min	>30 min	no	>30	>30

[1] Averages of several determinations. Negative results with boiled extracts used as controls were omitted from this table. >30 indicates no reaction after 30 min, negative.

[2] H_3PO_4, CH_3COOH, HCl, and H_2SO_4 are phosphoric, acetic, hydrochloric, and sulfuric acids, respectively. H_2O and EtOH are distilled water and 47.5% ethanol, respectively. Surfactant I = 0.005*M* of sodium alkyl aryl sulfonate. Surfactant II = saturated solution (about 0.002*M*) of sorbitan monostearate.

TABLE 4

Effect of different concentrations of hydrochloric acid (HCl) and of initial pH of infiltration solution on enzyme activity of treated cabbage shreds

Infiltration of solution[1]			Time required for positive reaction[2]				
Components of mixture	HCl conc. *N*	Initial pH of solution	Peroxidase seconds	Catalase		Acetyl esterase minutes	Phosphatase minutes
				Seconds	Foam ring		
H_2O	0.0	7.0	5	5	yes	2	6
H_2O + alkyl aryl sulfonate	0.0	6.6	3	10	yes	3	6
HCl + EtOH + alkyl aryl sulfonate	0.0005	4.1	2	15	yes	10	4
HCl + EtOH + alkyl aryl sulfonate	0.001 0.002 0.003	3.1	3	15	yes	10	4
HCl + EtOH + alkyl aryl sulfonate	0.04	1.8	15	60	no	>30	6
HCl + EtOH + alkyl aryl sulfonate	0.1	1.2	5 min	30 min	no	>30	>30

[1] H_2O, HCl, and EtOH are distilled water, hydrochloric acid, and 47.5% ethanol, respectively.

[2] Averages of several determinations. Negative results with boiled extracts, used as controls, were not included. >30 indicates no reaction after 30 min, negative.

TABLE 5

Effect of surface active agents (surfactants) on enzymes in infiltrated cabbage. The surfactants are listed in approximate order of effectiveness within each of the three groups

Trade name[1]	Surface active agent, chemical name	Conc. *M*	Infiltration medium[2]	pH of macerate	Time required for positive reaction[3]			
					Peroxidase	Catalase	Acetyl esterase, minutes	Phosphatase minutes
.....	Water	0	Water	5.7	3 sec	12 sec	3	4
Anionics[4]								
Utrawet D.S.	Na-decyl aryl sulfonate	0.005	Water	5.7	4 sec	5 sec	2	3
Utrawet D.S.	Na-decyl aryl sulfonate	0.005	HCl + EtOH	3.5	>30 min	>30 min	>30	>30
Nacconol	Na-alkyl aryl sulfonate	0.005	Water	5.8	2 sec	5 sec	1	10
Nacconol	Na-alkyl aryl sulfonate	0.005	HCl + EtOH	4.8	10 sec	>30 min	>30	>30
Tergitol 08	Na-sulfate of 2-ethyl hexanol-1	0.005	Water	5.8	5 sec	15 sec	7	5
Tergitol 08	Na-sulfate of 2-ethyl hexanol-1	0.005	HCl + EtOH	4.7	>30 min	>30 min	>30	>30
Alkanol SF	Na-tetrahydronaphthalein-sulfonate	0.005	Water	5.6	5 sec	20 sec	3	3
Alkanol SF	Na-tetrahydronaphthalein-sulfonate	0.005	HCl + EtOH	3.9	>30 min	10 min	>30	>30
Aerosol OT	Dioctyl ester of Na-sulfosuccinic acid	0.0023	Water	5.8	1 sec	15 sec	2	4
Aerosol OT	Dioctyl ester of Na-sulfosuccinic acid	0.0023	HCl + EtOH	4.2	>20 min	90 sec	>20	20
Arescarp 100	Monobutyl phenylphenol Na-sulfonate	0.005	Water	5.7	4 sec	10 sec	4	3
Arescarp 100	Monobutyl phenylphenol Na-sulfonate	0.005	HCl + EtOH	4.9	90 sec	60 sec	20	35
Nonionics								
Span 60 (62)	Sorbitan monostearate	<0.002	Water	5.5	5 sec	20 sec	2	3
Span 60 (62)	Sorbitan monostearate	<0.002 (saturated)	HCl + EtOH	4.3	180 sec	4 min	>30	>30
Igepal 880	Nonyl phenoxy polyoxyethylene ethanol	0.005	Water	5.7	6 sec	6 sec	6	4
Igepal 880	Nonyl phenoxy polyoxyethylene ethanol	0.005	HCl + EtOH	4.8	25 sec	60 sec	35	10
Tween 21	Polyoxyethylene sorbitan monolaurate	0.001	Water	5.6	4 sec	15 sec	3	8
Tween 21	Polyoxyethylene sorbitan monolaurate	0.001	HCl + EtOH	4.7	15 sec	90 sec	25	20
Triton 100	Alkylated aryl polyether alcohol	0.0005	Water	5.2	2 sec	12 sec	2	5
Triton 100	Alkylated aryl polyether alcohol	0.0005	HCl + EtOH	4.6	10 sec	75 sec	15	15
Cationics[4]								
Winthrop 379	Octadecyl guanidine hydrobromide	0.0025	Water	5.7	2 sec	17 sec	3	5
Winthrop 379	Octadecyl guanidine hydrobromide	0.0025	HCl + EtOH	4.6	8 sec	75 sec	>30	30
Emulsept	N-(2 acyloxyethyl) carbonamidomethyl pyridinium chloride	0.005	Water	5.9	10 sec	20 sec	5	3
Emulsept	N-(2 acyloxyethyl) carbonamidomethyl pyridinium chloride	0.005	HCl + EtOH	4.8	60 sec	90 sec	20	30
ATM 50	N-alkyl benzyl N-N-N trimethyl ammonium chloride	0.005	Water	5.9	2 sec	25 sec	2	3
ATM 50	N-alkyl benzyl N-N-N trimethyl ammonium chloride	0.005	HCl + EtOH	4.7	12 sec	60 sec	20	20
ROC	Alkyl dimethyl benzyl ammonium chloride	0.005	Water	5.2	2 sec	13 sec	3	4
ROC	Alkyl dimethyl benzyl ammonium chloride	0.005	HCl + EtOH	4.2	10 sec	60 sec	30	25

[1] Mention of trade names does not imply endorsement by the U. S. Department of Agriculture over others of a similar nature not mentioned.
[2] HCl + EtOH are 0.1*N* hydrochloric acid and 47.5% ethanol, respectively.
[3] >30 indicates no reaction after 30 min, negative.
[4] Two other commercial Na-alkyl aryl sulfonates as well as several nacconols were also tested. The results were similar to those presented in the table. An isopropyl naphthalene Na-sulfonate and a Na-lauryl sulfate were also tested and found comparable to compounds listed at the end of the anionics. A hexadecyl dimethyl benzylammonium chloride was found to fit at the end of the cationics.

apparent under our experimental conditions. The synergistic enhancement of enzyme inactivation when surface-active agents were combined with acid and ethanol may be due in part to increased diffusion rate of the reagents into the tissue.

As a group, the anionic surfactants were most effective in enhancing the synergistic action of acid and ethanol solutions, while the cationic compounds were the least effective. This observation is in agreement with studies showing that anionic detergents favor combination with cationic forms of protein on the acid side of the iso-electric point *(1, 15)*. This may result in precipitation of the protein-surfactant with lowered or destroyed enzyme activity.

SUMMARY

An effective procedure for inactivating enzymes by chemical means in cut plant tissue has been developed. Nearly complete inactivation of tested enzymes was observed in shredded cabbage and pieces of apple, carrot, and celery which had been infiltrated under vacuum with a mixture of acid, alcohol, and a surfactant.

None of these 3 components alone significantly inactivated the enzymes tested (catalase, peroxidase, acetyl esterase, phosphatase, and polyphenol oxidase) under the experimental conditions used.

Enzyme inactivation increased with decreasing pH of the infiltration solution. Strong mineral acids with a pH of 1.5 or lower in the infiltration mixture were best enzyme inactivators.

Anionic, cationic, and non-ionic surface-active compounds have been tested as additives to the acid-ethanol mixture used for enzyme inactivation. A number of the surfactants synergistically enhanced the enzyme-inactivating action of the acid-ethanol mixture.

The inactivation of enzymes increased with higher concentrations of the surface-active compound.

In general, the anionic surface-active compounds were most effective synergistic enzyme inhibitors in the acid-ethanol mixture, while the cationic compounds were least effective. It is difficult to assess from the available data the relation between chemical structure of the surfactant and its effectiveness in the synergistic inhibition of plant enzymes.

Of the enzymes tested, peroxidase appeared to be the most difficult to inactivate.

Acknowledgment

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The Pressure-Volume Relation in Film Packages During Heat-Processing[a]

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CONTAINERS for the commercial packaging of heat-processed foods are limited to those made from glass, or metals such as tin plate or aluminum. At present several plastic films with resistance to processing temperatures are available; these films are of interest to the food industry as possible alternatives to the conventional materials.

Heat-processing foods in plastic film containers presents a number of problems not encountered when containers made from conventional materials are used. Examples are:

a) Permeability of films to gases and vapors, and the effect of processing temperatures on these properties.

b) Possibility of extraction of film components into the food product during processing.

c) Effect of transmitted light on the food product.

d) Essential inertness of plastics. This may create additional problems. For example, tin and the special linings used in metal cans may react with food components or their breakdown products; such reactions are frequently beneficial to the appearance and flavor of the product. Other methods of achieving these desirable effects will have to be used for products heat-processed in plastics.

e) Effect of processing temperatures on the strength of films and heat-seals.

f) Internal pressure-volume relations in film packages during processing.

Some of the advantages and problems associated with the heat-processing of foods in film containers have been discussed by Hu *et al (4)* and by Nelson, Hu, and Steinberg *(6)*.

A detailed study of several of these problems has been conducted within the last two years in these laboratories. As part of these investigations, a method was developed for the estimation of the strength of completed packages and heat-seals before, during, and after heat-processing. Results were reported on several films *(3)*.

The present paper outlines investigations made on the internal pressure-volume relations in plastic containers, and includes:

a) Design of laboratory apparatus for the study of the pressure-volume relations.

b) Procedures used in these studies.

c) A record of observations made during processing on film packages filled with either water or a food product.

It is considered that the methods described may be suitable not only for use with heat-processed foods, but also for studying the pressure-volume relations in the "heat and eat" type of food packs; these packs have already found commercial applications *(1, 2, 7)*.

METHODS AND MATERIALS

Materials used. Films used were 0.001 in polyethylene terephthalate (polyester) film and 0.001 in 0.002 in, and 0.003 in high-density polyethylene blown tubing.

Construction of packages. Film packages were made from samples of cut sheet material by folding and heat-sealing along 2 edges to form a bag, and completing the end seal after the bags were filled. The heat-sealer used was a thermal impulse sealer (Model 13A, Vertrod Corporation, Brooklyn, N. Y.).

Estimation of expansion in volume. The expansion in volume of packages with increases in internal pressure was studied using the apparatus shown in Figure 1. The apparatus consists of a wide-mouthed glass jar fitted with a rubber stopper bored to accommodate an overflow tube, a burette, and a brass tube. One end of the brass tube is attached to the test bag by means of wing nuts, brass washers and rubber washers as described previously *(3)*. The other end of the tube is connected by pressure tubing to a mercury manometer and to a compressed air line. A valve is used for the control of the air pressure.

Estimations were made by connecting each bag to the brass tube positioned in the stopper, filling a measured volume of water into the bag, and closing after removing as much air as possible. To prevent leakage of water through the tubing during filling and closing operations, one end of the tube was

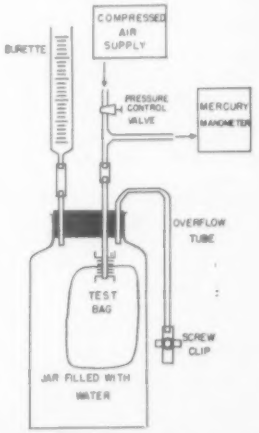

Figure 1. Apparatus used to measure the expansion in volume of film bags with increases in internal pressure.

[a] Contribution No. 368 from the Department of Food Technology.

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closed with a short length of rubber tubing clamped with a screw clip. The apparatus was then assembled, the jar filled with water from the burette, and all air removed from the jar through the overflow pipe. The overflow pipe was closed with a screw clip and the water level in the burette noted. The internal pressure in the bag was increased in small increments and the volume of water displaced from the jar into the burette observed, together with the pressure recorded on the manometer. The results reported were corrected to allow for the pressure exerted by the column of water in the burette.

Apparatus for heat-processing studies. The apparatus used for heat-processing test packs of products in film packages is shown diagrammatically in Figure 2. It is fitted with special attachments which permit estimation of the internal pressures developed in the packages. The apparatus consists of a standard Presto (Model 401B) domestic type pressure cooker fitted with a Weston dial thermometer (A) (range 50–300° F), a standard pressure gauge (B) (range 0–30 in Hg), and a pressure control valve. The valve stem (D) fixed to the lid of the cooker, has a socket in the upper end which fits a ball in the valve cover (C). Heat-processing in steam at 212° F may be carried out with the valve cover removed, and processing at higher temperatures under steam pressure by using valve covers of suitable weights. The attachments for the estimation

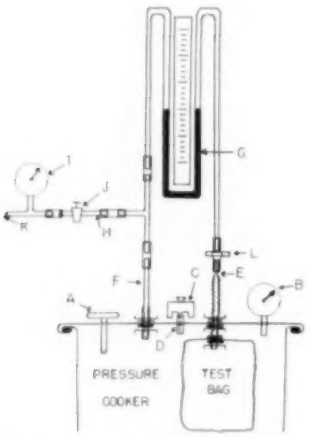

Figure 2. Apparatus used for heat-processing studies.

of internal bag pressures consist of a mercury manometer (G) attached by pressure tubing to two brass tubes (E, F). The brass tubes are fixed to the lid of the cooker by wing nuts, brass washers and Teflon washers. The test bag is sealed to the lower end of tube E. The brass washer outside the bag is permanently soldered to the tube to prevent escape of gases and vapors from the bag along the thread. Tube E is threaded over the full length, but a short length of the thread on the upper end is removed permitting the wing nut used to clamp the tube to the cooker lid to be removed from this end. Provision is made for cooling the cooker under super-imposed air pressure from a compressed air source (K) connected by pressure tubing and a T-piece inlet to tube F. Air pressure is controlled by a pressure gauge (I) and a control valve (J).

Procedure for pressure estimation. Each bag was attached to tube E as described earlier and the other end of the tube was closed with a piece of rubber tubing and a screw clip (L). The bag was filled, closed and the end of the tube inside the bag temporarily sealed by finger pressure. After removing the piece of rubber tubing, the tube E was attached to the cooker lid by the washers and wing nuts, and the piece of rubber tubing replaced. The apparatus was then assembled and the manometer connections secured. The internal pressure developed in the bag during the heat-processing was determined from the difference in height of the mercury levels of the manometer.

Estimation of enclosed air in packages. To study the effect of the volume of enclosed air on the internal pressures it was necessary to estimate the volume of air present.

Procedure. At the completion of the heat-process during which the internal pressure was recorded, the bag with the tube attached was removed from the cooker by a reversal of the assembly procedure outlined above. Volume of enclosed air was then estimated by puncturing the bag under water and collecting the gas in a gas burette. Volume of air in the burette was read at room temperature (70–75° F) and atmospheric pressure.

Test packs of food products. Three test packs of peas and one of mixed peas and carrots were prepared. The food material was obtained in frozen form and allowed to thaw before the bags were filled. Two of the test packs of peas were prepared in 5 in x 3 in bags with 50 g of peas and 60 ml of 2% brine containing 2% sugar. The third test pack was prepared in 4 in x 3 in bags using 30 g of peas and 25 ml of the brine solution. Mixed peas and carrots were prepared in 5 in x 3 in bags with 45 g of mixed vegetables and 45 ml of 2% brine. Care was taken to expel as much air as possible from the bags before closing.

RESULTS AND DISCUSSION

Pressures which develop inside sealed containers during heat-processing arise from:

a) Increase in the vapor pressure of the water in the processed food, with increasing temperature.

b) Increase in the pressure of air in the headspace, with increasing temperature.

c) Release of additional air from the product, due to a decrease in gas solubility with increasing temperature.

d) Thermal expansion of the food product itself.

In rigid containers packed with foods to be heat-processed a partially evacuated headspace is provided. One reason for this procedure is to reduce the strain on the container during the heat-process. Tin-plate containers are not perfectly rigid and will increase in volume slightly as the internal pressure increases, resulting in further reduction of the final internal pressure. Nevertheless, internal pressures in tin-plate containers are relatively high *(5, 8)*.

Containers made from plastic films have a much lower resistance to internal pressure than those made from tin plate or glass. The internal pressures developed in film packages during heating are therefore of considerable importance. Fortunately, processing in flexible containers results in much lower internal pressures than those developed in cans. This is due primarily to the greater expansion in volume of film containers *(9)*. This effect, characteristic of processing in plastic, is evident from the results shown below.

Expansion in volume of bags. The expansion in volume with increases in internal pressure was estimated on bags made from 0.001 in polyester and 0.001 in and 0.003 in high-density polyethylene films. The bags (4 in x 3 in) were filled with 50 ml of water and closed after the removal of air. Estimations were made on several bags from each film. Only slight variation was found between bags made from the same film. The results, Figure 3, show that 4 in x 3 in bags made from the films and filled with 50 ml water increased in volume by an additional 55–65 ml at an

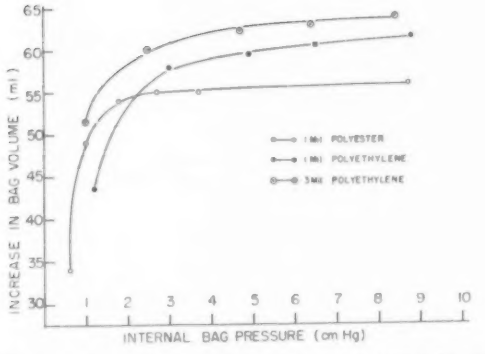

Figure 3. Increase in volume of 4 in x 3 in bags filled with 50 ml water with increases in internal pressure.

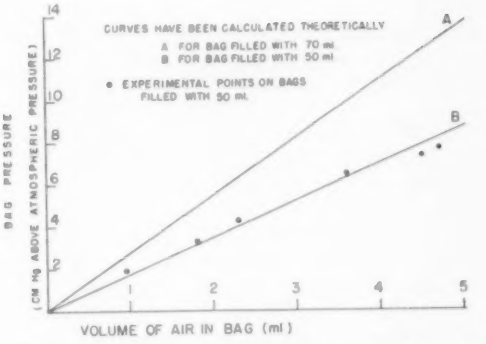
CURVES HAVE BEEN CALCULATED THEORETICALLY
A FOR BAG FILLED WITH 70 ml
B FOR BAG FILLED WITH 50 ml
EXPERIMENTAL POINTS ON BAGS FILLED WITH 50 ml
BAG PRESSURE (cm Hg ABOVE ATMOSPHERIC PRESSURE)
VOLUME OF AIR IN BAG (ml)

Figure 4. Effect of the volume of air enclosed in 4 in x 3 in bags made from 0.001 in polyester film on the internal pressures developed during processing at 212° F.

internal pressure of 5–10 cm Hg. With bags of this size, the maximum volume of a liquid product which can be enclosed without risks of spillage is approximately 70 ml. Results show that even if such bags are filled with 70 ml of water, they will still expand in volume by 35–45 ml. This relatively large expansion would prevent the development of high internal pressures during heating.

Internal pressure during processing at 212° F. If the total internal volume of a bag and the volume of enclosed air are known, the internal pressure developed during heat-processing in a bag filled with a measured volume of water may be calculated from the gas law equation:

$$\frac{P_1V_1}{T_1} = \frac{P_2V_2}{T_2}$$

where:

P_1 — initial pressure in bag taken as 76.0 cm Hg.

V_1 — volume of enclosed air at temperature T_1 and pressure P_1.

T_1 — initial temperature of bag contents °R.

P_2 — partial pressure of water at temperature T_2 plus partial pressure of enclosed air at temperature T_2 and volume V_2.

V_2 — volume of bag at pressure P_2, and temperature T_2.

T_2 — temperature of heat-process °R.

The calculated curves showing relation between internal bag pressure and volume of initial air for 4 in x 3 in bags filled with 50 and 70 ml of water and processed at 212° F are shown in Figure 4. In these calculations the initial temperature was taken as 70° F and final volume of the bags as 105 ml. No allowance was made for increase in the specific volume of water since it is negligible compared with increase in volume of the bags.

Maximum internal pressures in 4 in x 3 in bags made from polyester film, and filled with 50 ml of water and closed without complete removal of air were determined during processing at 212° F. At the end of the process the volume of enclosed air in the bags was determined. Experimental values, also shown in Figure 4, are in good agreement with the calculated curve.

Results show that pressures which develop in film packages filled with water and heat-processed at 212° F are influenced by the volume of air enclosed and by the amount of water present. Data of this nature, together with data on the strength of bags at the process temperature, would allow the calculation of the optimum fill of containers.

Internal pressure during processing at temperatures above 212° F. Conventional retorts for the heat-processing of foods at temperatures in excess of 212° F utilize a high pressure steam supply. The steam pressure in the retorts is controlled to give the required temperature. Pressure in the container is equal to the vapor pressure of water and to the expansion of the product and of the enclosed air. Because of the lag in the temperature rise of the product, vapor pressure of water in the container rises more slowly than the retort pressure. The maximum internal pressure is attained when the product reaches the retort temperature. It is then equal to the retort pressure plus the pressure due to expansion of product and of entrapped air.

Of more interest in package design, however, is the differential pressure ΔP, which is defined as follows:

$$\Delta P = P_I - P_R$$

where: P_I — internal container pressure at a given time

P_R — retort pressure at the same time.

During the initial stages of heat-processing the value for ΔP is relatively small, and may even be negative. When the product reaches retort temperature and P_I attains its maximum value, the vapor pressure of the water in the container is equal to P_R. ΔP is due entirely to expansion of the product and of the enclosed air. The maximum ΔP, however, occurs during the cooling period because the retort pressure falls more rapidly than the pressure inside the container. To minimize the resulting strains on containers during cooling, retorts are generally cooled under superimposed air pressure.

Preliminary trials made with film packages filled with water and processed at 250° F in a normal steam retort resulted in very high proportion of burst packages, even when cooling was done under superimposed air pressure. This was probably due to difficulties in maintaining the retort pressure at a constant level.

Observations made using the conventional type retort indicated the desirability of obtaining more complete information on the internal pressure of bags during heat-processing. This was obtained on 4 in x 3 in polyester bags filled with 50 ml of water using the modified cooker described. In one experiment the bag was attached to the differential manometer and processed for a short time in steam at 212° F, the steam valve cover was then placed on the valve and the bag processed at 232° F in steam at 8 psig. The cooker was allowed to cool in air at the end of the process. The differential pressure in the bag was recorded at intervals during the processing and cooling period. The results (Figure 5) show a rise in ΔP to approximately 5 cm Hg during the process at 212° F and 232° F and a large increase to nearly 12 cm Hg as the cooker pressure fell to atmospheric pressure (Figure 5, part A). ΔP then began to decrease (part B). Similar results were obtained during processing at 240° F and 250° F, but the bags in each trial burst before the cooker pressure reached atmospheric pressure. These results clearly indicate that the maximum differential pressure is developed during the cooling operation.

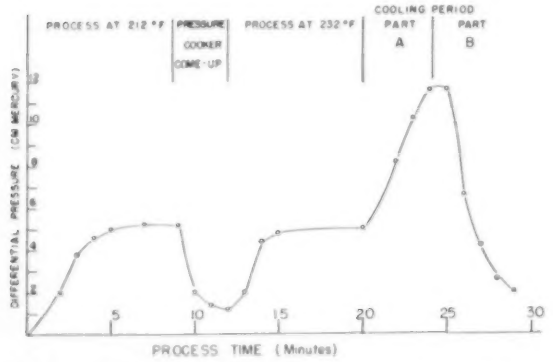

Figure 5. Differential pressure in a 4 in x 3 in bag filled with 50 ml water during processing and cooling.

In all subsequent processing trials at temperatures above 212° F, the cooker was cooled using superimposed air pressure. The procedure used to process a batch of bags filled with water or a food product was to attach a bag of similar dimensions and filled with the same material as those comprising the batch, to the differential manometer (Figure 2). The heat-process was then completed. When the cooling operation was begun, enough compressed air was admitted to the cooker to keep the differential pressure at a low level. Rapid cooling of the cooker under running tap water was possible using this method. Thus, the bag attached to the manometer serves as an indicator of the differential pressure in the batch, and close control of the cooker pressure with compressed air is possible during cooling.

A series of test packs was processed in the cooker at temperatures above 212° F using superimposed air pressure during cooling under tap water as outlined above. The maximum observed differential pressures developed in the control bag are recorded in Table 1.

TABLE 1

Maximum differential pressures observed in bags made from 100 gauge polyester film during processing

Bag size (in)	Contents	Process temperature (° F)	Differential max. pressure (cm Hg)
4 x 3	50 ml water	232	5.6
4 x 3	50 ml water	232	5.0
4 x 3	50 ml water	250	1.0
4 x 3	50 ml water	250	0.8
4 x 3	70 ml water	250	1.4
6¼ x 5	450 ml water	250	5.8
6½ x 5½	450 ml water	250	4.8
5 x 3	50 g peas and 60 ml brine	240	5.2
5 x 3	50 g peas and 60 ml brine	240	6.8
5 x 3	45 g mixed peas and carrots and 45 ml brine	240	6.0
4 x 3	30 g peas and 25 ml brine	240	5.0

The test packs were examined after processing for bag failures, and no failures were found with bags made from polyester film. In several of the trials, bags made from 0.001 in 0.002 in and 0.003 in high-density polyethylene film were included, but a high proportion of failures were found with these films. Most failures occurred in the area adjacent to the seals or in the seals themselves. A more detailed discussion of the mechanism of failure and its dependence on sealing conditions has been given previously *(3)*.

These results agree with the previously observed lowering of strength of bags made from polyolefin films when the bags are exposed to strains at high temperatures, which may result in failures even prior to the cooling period *(3)*. It appears logical that if these materials are to be used for heat-processing an almost complete elimination of enclosed air would be necessary.

SUMMARY AND CONCLUSIONS

Suitable laboratory apparatus and procedures for the study of the pressure-volume relation in film packages during heat-processing have been described.

The expansion in volume with increases in internal pressure was studied on packages made from several films. Results showed that the expansion in volume of the packages was sufficient to prevent high internal pressures developing during heat-processing. Internal pressures were shown to be related to the amount of material and enclosed air in packages of given dimensions when processed at a given temperature. Excellent agreement between calculated and estimated pressures was obtained on packages filled with water and processed at 212° F. The possible use of this

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technique as a guide for choosing the correct fill-in weights for packages to be heat-processed has been indicated.

Differential pressures in film containers filled with water and several food products during processing at temperatures above 212° F under steam pressure were found to be relatively small except during the cooling operation. Cooling under superimposed air pressure was found to avoid the development of high pressures. The use of a package attached to a manometer as an indicator of the correct air pressure to be maintained during cooling has been described.

Acknowledgment

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Control of Oxidative Changes in Irradiated Hams[a]

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(Manuscript received September 18, 1959)

IRRADIATION OF CURED MEATS leads to certain oxidative changes which alter their flavor and color. Although the flavor changes are not particularly objectionable *(12)*, it is desirable to control them as much as possible.

Many investigators have studied possible causes of irradiated off-flavors. Presence of oxygen during irradiation results in an increased amount of oxidative changes in meats *(7, 12)*. Addition of antioxidants such as ascorbic acid and liquid smoke helps control these oxidative changes *(8)*. Ascorbic acid has been used successfully to protect unirradiated meats against oxidative rancidity and meat discoloration *(2, 15, 17)*. Liquid smoke and ascorbic acid act synergistically to protect unirradiated ground pork against rancidity *(16)*, and to protect hams and bologna against radiation-induced changes in pigment and odor.

The ultimate purpose of radiation is to sterilize the cured ham and prolong its shelf life without need for refrigeration. Heat must still be applied to develop the cured meat color. The question arises as to when is the best time to heat the hams for getting the maximum color development. Is there an advantage to waiting until after irradiation before heating the hams; and if so, how long a time interval should lapse between the irradiation and heating processes?

Morgan and Reed suggest that heating hams after irradiation would be more effective from the standpoint of total sterilization *(10)*. Cain *et al* have shown that bacterial survival is dependent upon both irradiation dose and temperature whereas irradiation flavors are dependent only upon the irradiation dose *(3)*.

This paper reports data from experiments planned with two objectives in mind: first, to determine whether hams should be heated prior to or at some time interval after irradiation; second, to investigate the use of ascorbic acid and liquid smoke in protecting irradiated hams from oxidative changes.

MATERIALS AND METHODS

Approximately 40 lb of fresh pork hams were purchased from local markets, ground medium-fine in an electric food chopper, mixed thoroughly with curing salts, sealed in 307 x 113 C-enamel cans[c] (175 g per can) and frozen. Curing salts were 2% NaCl and 200 ppm nitrite as $NaNO_2$. Variations in chemical additives were (a) control and (b) 0.1% ascorbate plus 0.05% liquid smoke.[d] Frozen samples were packed in dry ice and shipped by air express to the Argonne National Laboratories where they received two million rep[e] gamma rays at ambient temperatures. Part of the samples were heated for color development prior to shipping and the rest were heated immediately after their return or at time intervals of 15 days and 32 days after irradiation. Samples were heated in an autoclave with streaming steam until internal temperatures reached 70° C. The unirradiated controls received the same treatment, in regard to handling, as the irradiated samples. Storage of the samples at room temperature was for various periods as indicated in the tables. Analytical data were obtained on pigment loss, fat oxidation, sulfhydryl groups, free nitrite, drip, pH, and odor acceptance.

Representative extinction curves for the cured meat pigment show a broad absorption peak at 570 mμ and very low absorption at wave lengths above 650 mμ. Erdman and Watts have shown the absorption peak to shift from 570 mμ to 540 mμ and finally disappear altogether when the pigment is oxidized *(6)*. Absorption ratios at 570/650 mμ will change from 2.56, which represents a completely developed cured meat pigment, to 1.56 when the pigment is oxidized to the ferric form without porphyrin ring destruction. The absorption ratios at 540/500 mμ change from 1.05 to 0.95 when the porphyrin ring is altered. Reflectance measurements at these wavelengths were therefore used to determine the kind and extent of pigment loss during irradiation.

Fat oxidation was measured with thiobarbituric acid (TBA) reagent according to the method of Turner *et al (14)*. The TBA value has been defined in a previous article *(9)*. Excess nitrite interfers with the production of the pink TBA-color complex *(13)*. However, this method can be considered valid for hams when the concentration of nitrite is less than 100 ppm. Hougham and Watts have shown a rapid loss of nitrite in hams during storage *(9)*.

Sulfhydryl groups were determined qualitatively with nitroprusside as described by Erdman and Watts *(4)*. Nitrites were measured quantitatively by the A.O.A.C. official method *(1)*.

The pH was measured on a Beckman pH meter. Drip was measured as the volume of drained liquor from a freshly-opened can. The drained liquor was added back to the samples when they were ground and mixed for analysis.

A modified hedonic scale described by Peryam and Pilgrim *(11)* was used to determine odor preference. Odor was judged by 14 to 17 volunteers who judged each sample by checking one of the following criteria: *dislike extremely, dislike moderately, neither like nor dislike, like moderately,* and *like extremely*. The ranks were then scored 1, 3, 5, 7, and 9, respectively. Judging was done in a well ventilated room that was not used for other purposes. Samples receiving the heat treatment prior to and immediately after irradiation were judged separately from the other samples, thus effectively reducing to four the number of samples handled by each judge at any one judging period.

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[a] This paper reports research undertaken by Florida State University in cooperation with the Quartermaster Food and Container Institute for the Armed Forces, and has been assigned number 1040 in the series of papers approved for publication.

[b] Present address: Iowa State University, Ames, Iowa.

[c] The American Can Company provided the 307 x 113 C-enamel cans.

[d] The liquid smoke was provided by Fermenich and Co., Geneva, Switzerland.

[e] The approved term, it is recognized, is now rad (100 ergs/g) but at the time of the experiments the term rep was sanctioned as the unit of dosage.

RESULTS AND DISCUSSION

Unirradiated hams spoiled when stored beyond one month and so no data are available for unirradiated hams kept several months.

Pigment loss. A change in the 570/650 absorption ratios indicated a definite loss of the cured meat pigment when the hams were exposed to ionizing radiations, Table 1. A smoke-ascorbate combination offered slight protection to the hams that were heated prior to or immediately after irradiation. By waiting 2 to 4 weeks after irradiation to heat the hams, there was considerably less pigment formation and the addition of smoke and ascorbate resulted in even less development of color.

Irradiated hams stored up to 265 days had higher 570/650 mmc ratios than did hams analyzed soon after irradiation, but there was an apparent loss of pigment in the unirradiated hams during storage up to a month. Erdman and Watts showed a preheated irradiated cured meat sample lost all of its pink color within 2 weeks of storage *(4)*. In this set of data, however, preheated irradiated hams had the best color rating of all samples after 265 days in storage. The rapid loss of nitrite during storage would explain, in part, the poor development of the pink color in hams held 15 and 32 days after irradiation before heating.

TABLE 1

Effect of radiation and heating on the 570/650 mmc absorption ratios

Heating period	Storage time in days	No smoke or ascorbate		Smoke plus ascorbate	
		2×10^6 rep	No rep	2×10^6 rep	No rep
Prior to irradiation	6	2.20	2.56	2.38	.....[1]
	34–36	2.36	2.46	2.26	2.55
	265	2.43	.....	2.32	.....
Immediately after irradiation	6	2.25	2.50	2.36	2.46
	34–36	2.26	2.36	2.30	2.42
	265	2.32	.....	2.28	.....
Fifteen days after irradiation	19	2.06	2.36	1.98	.....
	34–36	2.16	2.06	1.96	.....
	265	2.23	.....	2.16	.....
Thirty-two days after irradiation	34–36	1.99	.....	2.03	.....
	265	2.15	.....	2.20	.....

[1] Sample lost to experiment. Other blank spaces indicate spoiled samples.

There was no difference in the 540/500 mmc absorption ratios between irradiated hams and corresponding unirradiated hams. However, addition of smoke and ascorbate had some unexplained effect on this absorption ratio. Ratios for hams without smoke and ascorbate ranged from 1.04 to 1.00 during storage and for hams with smoke and ascorbate from 1.03 to 0.97. Investigation showed that the presence of smoke in freshly prepared hams will cause a change in ratios from 1.06 to 1.05. This is an insufficient change to account for the sizeable decrease in the absorption ratios on the basis of added light absorption by the liquid smoke itself. Analysis of variance gave a mean square = .0039 for the treatment (df = 1) and .0003 for the individual variation (df = 29); F = 13.00; p = .005. This shows a very high probability of getting lower 540/500 mmc absorption ratios when smoke and ascorbate are added. It is not known how the smoke-ascorbate additives effects the porphyrin structure of the meat pigment.

Sulfhydryl and nitrites. Irradiation of the hams produced a difference in the visual ratings for free SH groups and free nitrite. There was a slight increase in free SH groups in the hams held 15 and 32 days before heating. The smoke and ascorbate made no differtnce in the ratings for SH groups. There was a rapid loss of SH groups during storage. This agrees with observations previously made by Erdman and Watts *(4)*.

Nitrite concentration decreased very rapidly during storage and there was evidence that the rate of loss of nitrite in unirradiated hams was greater than the rate of loss in irradiated hams. Irradiated hams showed a trace of residual nitrite up to one month storage but the unirradiated samples gave no color test for nitrite at one month storage.

The presence of smoke and ascorbate caused a decided loss of free nitrite in hams. At 6 days' storage, the color rating for hams containing smoke and ascorbate showed only a trace of nitrite. In those hams containing no smoke or ascorbate, the color rating was maximum for the nitrite color test. This loss of nitrite can be ascribed to the presence of ascorbic acid in the hams.

Fat oxidation. TBA values for all irradiated hams remained low throughout the storage periods. The average mean value for those hams containing smoke and ascorbate was 0.28 ± .01 and for those with no smoke and ascorbate was 0.33 ± .02. A TBA value typical of rancid meats is greater than 0.50 *(13)*. The low TBA values could not be due to interference from free nitrite because of the rapid loss of nitrite during storage. It is more likely that the nitrosomyoglobin in cured meat is unable to catalyze the oxidation of fats. Heating the hams at various periods before and after irradiation made no differences in the TBA values. Adding smoke and ascorbate probably gave protection to the hams against the slight amount of irradiation induced fat oxidation.

Sensory tests. Judges showed a decided preference for irradiated hams containing liquid smoke and ascorbate, Table 2. Samples tested at 265 days were

TABLE 2

Effect of radiation and heating on odor acceptance of stored samples

Heating period	265 days storage		313 days storage	
	No smoke or ascorbate	Smoke plus ascorbate	No smoke or ascorbate	Smoke and ascorbate
Prior to irradiation	3.8	4.3	3.1	5.3
Immediately after irradiation	3.0	4.3	2.3	5.7
Fifteen days after irradiation	3.5	4.4	3.4	5.1
Thirty-two days after irradiation	2.8	4.8	3.7	6.0
mean =	3.1	4.4	3.2	5.5
n =	16	17	14	14

Irradiation dose = 2×10^6 rep for all samples.

ground and mixed for chemical analysis before making the sensory tests. Samples tested at 313 days were opened within a half hour before testing and were not ground but were trimmed of all surface discoloration. In both testing periods, hams containing smoke and ascorbate were significantly ($p = .01$) more acceptable to the panel of judges.

In irradiated hams without smoke and ascorbate, a t-test showed no significant difference between hams heated prior to irradiation and those heated immediately after irradiation. This means that the smoke-ascorbate combination was the major factor in masking the irradiated odor and in increasing its acceptability to the group used in these sensory tests. No doubt the odor of the smoke and not the presence of the ascorbate made the difference.

Drip loss and pH. There was no noticeable difference in pH due to the ionizing radiations, to the presence of smoke and ascorbate, or during storage. Drip losses declined during storage of the samples, Table 3. Denaturation of the protein and subsequent formation of cross links would result in a change in the water-binding capacity of the hams. Higher drip losses occurred if hams were held several days after irradiation before heating.

TABLE 3

Drip losses during storage of irradiated samples[1]

Heating period	Storage time in days	% Drip during days of storage	
		No smoke and ascorbate	Smoke plus ascorbate
		%	%
Prior to irradiation	6	13.0	12.8
	30	14.8	14.8
	137	13.1	10.9
	265	12.3	11.1
Immediately after irradiation	6	13.4	12.8
	30	10.3	10.9
	137	9.7	10.9
	265	11.2	9.0
Fifteen days after irradiation	19	19.5	17.8
	34	18.9	17.8
	137	15.4	14.3
	265	15.1	13.8
Thirty two days after irradiation	34	21.7	20.6
	137	17.1	17.1
	265	16.6	15.4

[1] Radiation dose = 2×10^6 rep.

SUMMARY

A combination of heat and irradiation for the production of a good cured pork product with a long shelf life has been studied. Although there is no apparent advantage to heating hams prior to or immediately after irradiation, it is apparent that no long interval of time should lapse between the two events.

A smoke and ascorbate combination showed some antioxidant behavior. The smoke helped to mask the off-odors. The presence of ascorbate caused an unnecessary loss of nitrite and SH which led to an undesirable loss of pigment.

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Chemical and Color Changes in Canned Tomato Ketchup

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(Manuscript received August 13, 1959)

CHEMICAL AND COLOR CHANGES in tomato ketchup are of prime interest to processors. Various investigators have studied color *(2, 3, 7, 9)*, ascorbic acid content *(5)*, tomato content *(1, 6)*, preservatives *(8)*, and consistency *(11)* as factors influencing the quality of tomato ketchup.

Davis and Kefford *(3)* investigated "black neck" in tomato sauce and concluded that the iron-tannin-oxygen reaction is one of the mechanisms responsible for color deterioration. Ogle and Kramer *(7)* reported on the browning of ketchup in bottle necks during storage. However, not much is known about the storage stability of ketchup in cans, even though this product is widely used in institutional feeding.

One of the programs of the Department of Food Science and Technology at the Davis campus concerns factors influencing storage stability of processed foods. The interest shown by canners in the stability of canned ketchup gave impetus to a study on the effect of storage temperature and time on chemical and color changes in canned tomato ketchup. Several objective tests for evaluating storage changes are presented.

EXPERIMENTAL

Tomatoes. Pearson tomatoes at canning ripeness were used for making ketchup. Chemical analyses of the tomatoes were made. The Agtron E reading was 45, Brix reading 5.95 at 20° C, titratable acidity as citric acid 0.472%, pH 4.25, and ascorbic acid content 20.2 mg/100 g.

Tomato ketchup. The tomatoes were washed, sorted, and passed through a Reitz Thermopulper where the tomatoes were heated to 200° F in 10 sec. The output of the thermopulper was 5 gal per min. The product was passed through a pulper with a 0.030 in screen. Forty gallons of the tomato puree was evaporated in a Krenz Evaporator at 106° F under 29 in vacuum to 19° Brix. Then, 30 lb of sucrose, 5 lb of canners' salt, 6.4 oz of mixed ketchup spices (William J. Stange Company), and 1.3 gal of 100 grain vinegar were added. This product was mixed well, brought to a boil, filled hot into No. 2½ cans (¾–½ in head space), sealed, heated in a rotary cooker for 5 min at 212° F, and rapidly cooled in water. Eighty-five cans of tomato ketchup were obtained. Chemical analyses of the ketchup were made. The Brix reading was 30.0° at 20° C, titratable acidity as citric acid 1.31%, pH 3.70, and ascorbic acid 15.6 mg/100 g. The cans were stored at temperatures varying from 20° C (68° F) to 55° C (131° F) until needed for chemical and color studies.

ANALYTICAL METHODS

Serum color. Twenty grams of the ketchup sample was diluted with distilled water to 100 ml in a volumetric flask. The mixture was centrifuged to remove the insoluble solids. Then, 20 ml of the centrifugate was mixed with 2 ml of a freshly prepared and filtered solution of 10% (w/v) Pectinol R-10 (Rohm and Haas Co., Philadelphia, Pa.). The mixture was kept at 40° C (104° F) for 1 to 2 hours, treated with 0.2 g of Supercel, and filtered through a Whatman No. 2 filter paper. The absorbance of the filtrate was measured in a Klett-Summerson photoelectric colorimeter with a blue filter. A blank was run with distilled water instead of the centrifugate.

Amino nitrogen. The ninhydrin colorimetric method described previously *(4)* was modified slightly for this experiment. Twenty grams of the ketchup sample was diluted with distilled water to 100 ml. Five milliliters of the solution was diluted to 100 ml with distilled water and then filtered. Two milliliters of the filtrate was pipetted into a 50 ml volumetric flask with a glass stopper. To this was added 5 ml of KCN-pyridine reagent (2 ml of 0.01M KCN solution diluted to 100 ml with ammonia-free pyridine, which was prepared by shaking 100 ml pyridine with 1 g of permutite for 20 min) and 5 ml of 80% phenol solution (80 g of phenol dissolved in 20 ml of absolute alcohol shaken with 1 g of permutite for 20 min). The flask was stoppered and heated in a boiling-water bath for 3 min. After partial cooling, 1 ml of 5% ninhydrin solution in absolute alcohol (w/v) was added. The mixture was heated in a boiling-water bath for exactly 5 min, counting from the time the water started to boil. The solution was cooled in a cold-water bath and made up to 50 ml with 60% ethyl alcohol. The absorbance of the blue solution was measured in a Klett-Summerson photoelectric colorimeter with a green filter. A reagent blank was run, using distilled water instead of the sample. A standard curve was constructed, using glutamic acid concentrations up to 150 mcg per ml of sample. The results were expressed as mg amino nitrogen per 100 g of ketchup.

Folin-Denis test. The modified Folin-Denis test *(4)* was used to determine tannin-like substances in ketchup. Twenty grams of ketchup was diluted with distilled water to 100 ml in a volumetric flask. Then, 2 ml of the supernatant was mixed with 50 ml of distilled water, 5 ml of Folin-Denis reagent, and 20 ml of 1M Na_2CO_3 solution. The mixture was diluted to 100 ml and allowed to stand for 90 min. The absorbance of the blue colored solution was measured in a Klett-Summerson photoelectric colorimeter, using a No. 66 red filter. It was sometimes necessary to centrifuge the solution for 5 min to remove the white precipitate. A standard curve for cp tannic acid was constructed with concentrations ranging from 0.05 to 1.00 mg of tannic acid per 100 ml of reaction mixture. The results were reported as mg of tannic acid per 100 g of ketchup.

Titratable acidity. Ten grams of the ketchup sample was diluted with 150 ml of distilled water and then titrated with 0.1N NaOH to pH 8.0, using a Beckman glass electrode pH meter. Results were reported as per cent citric acid.

Lycopene and beta-carotene. Twenty grams of the ketchup sample was diluted to 100 ml with distilled water and centrifuged. The residue was resuspended in 100 ml of water and recentrifuged. The residue was extracted several times with a total of 100 ml of acetone. The carotenoids were then transferred to 75 ml of petroleum ether and washed with 50 ml of water. The petroleum ether extract was treated with anhydrous Na_2SO_4 and made up to 100 ml with petroleum ether. A 5-ml aliquot of the extract was used for chromatography on a column (⅞ in in diameter and 4 in tall) made with equal weights of MgO and Supercel. The beta-carotene was eluted from the column with an acetone and petroleum ether mixture (10:90 v/v). The xanthophylls were mechanically scraped off the top of the column. The lycopene was then eluted from the column with a mixture containing 5% ethyl alcohol and 95% petroleum ether. The pigments were measured in a Beckman DU spectrophotometer at 472 and 502 mmc for lycopene, and 450 and 478 mmc for beta-carotene.

Color. The color grades of the tomato ketchups were determined according to the U.S.D.A. Munsell color disc method *(12)*. The color of the ketchup was determined by comparing it with that produced by spinning a Munsell disc for fancy grade ketchup, composed of the following colors: red 65% (5R 2.6/13, glossy finish), yellow 21% (2.5 YR 5/12, glossy finish), black 7% (N1, glossy finish) and gray 7% (N2, mat finish). Ketchup samples with color grades of 21–25 points were graded as fancy, 17–20 as standard and 0–16 as substandard.

RESULTS

Serum color. When the tomato ketchup samples were stored at temperatures higher than 20° C (68° F), the ketchup serum darkened rapidly. Results shown in Figure 1 indicate the amount of serum-color darkening at different temperatures. The serum color of the sample stored at 20° C (68° F) for 408 days darkened very slightly. The rate and extent of darkening increased as storage temperature rose. Serum color might

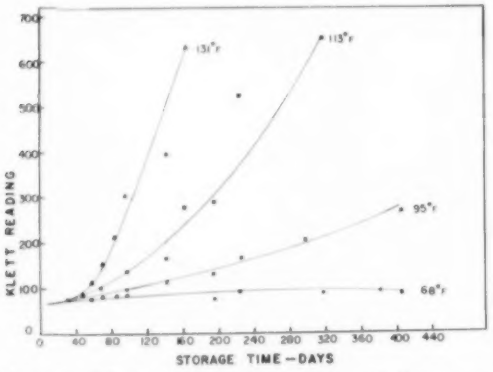

Figure 1. Effect of storage temperature and time on serum color of canned tomato ketchup.

therefore be used as a test of ketchup quality. If one considers a Klett reading of 100 to be maximum for good-quality ketchup, then the maximum storage time for canned ketchup will be 100 days at 35° C (95° F). Data reported here indicate the importance of storage temperature to product stability. For better quality retention it is desirable to store the canned ketchup at 20° C (68° F) or lower.

Amino nitrogen. Amino acids and proteins are related to flavor acceptance of tomato products *(4)*. A measurement of amino nitrogen might be used as a criterion of tomato ketchup quality. When ketchup was stored at temperatures above 20° C (68° F), the amino nitrogen content decreased rapidly. Figure 2 indicates the trend of such changes. The rate of decrease in amino nitrogen was far more rapid for the sample stored at 55° C (131° F) than for those stored at lower temperatures. At 20° C (68° F), it took 408 days for a decrease in amino nitrogen from 80 mg/100 g to 74.5 mg/100 g.

The decrease in amino nitrogen content during storage may be considered undesirable. This can be avoided by storing the ketchup in the lower temperature range. In another experiment, it was found that storage at 0° C (32° F) was superior to storage at 20° C (68° F) for amino nitrogen retention. However, for practical purposes, a storage temperature of 20° C (68° F) would be an acceptable compromise for a low storage cost with satisfactory quality retention.

Folin-Denis test. Storage changes in tomato ketchup may also be determined by the Folin-Denis test which measured tannin-like substances containing aromatic hydroxyl groups. This test is indicative of changes caused by high temperature, long storage, or both. Results shown in Figure 3 indicate a gradual liberation during storage of substances which react with the Folin-Denis reagent. A trend similar to that of serum color

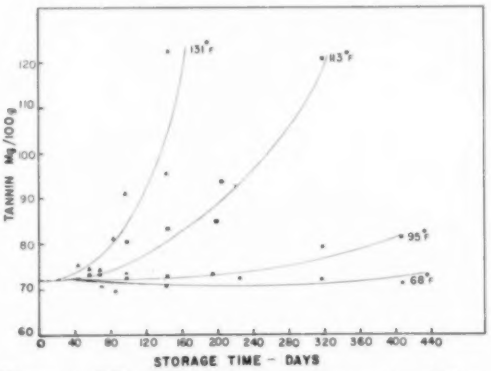

Figure 3. Effect of storage temperature on tannin-like substances in canned tomato ketchup.

darkening was obtained. It is apparent that the serum-color and the Folin-Denis tests have similar temperature coefficients.

It is important to point out that the Folin-Denis test is not specific to tannin-like substances. Ascorbic acid, tyrosine, stannous ion and other substances may also react with the reagent to produce a blue color. However, this test is useful for quality control purposes because improperly stored samples give high values.

Titratable acidity. When canned ketchups were stored at high temperatures, an undesirable metallic flavor and even hydrogen swelling occurred. This was found to be accompanied by an increase in titratable acidity. Results shown in Figure 4 indi-

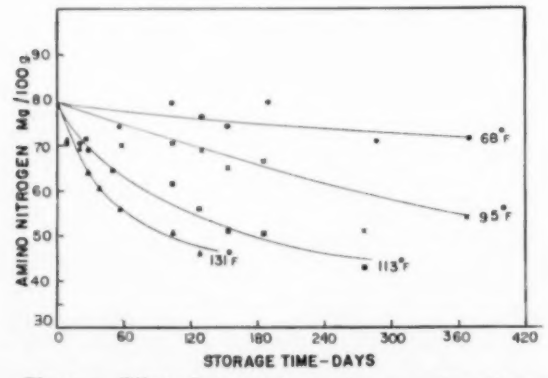

Figure 2. Effect of storage temperature on amino nitrogen content in canned tomato ketchup.

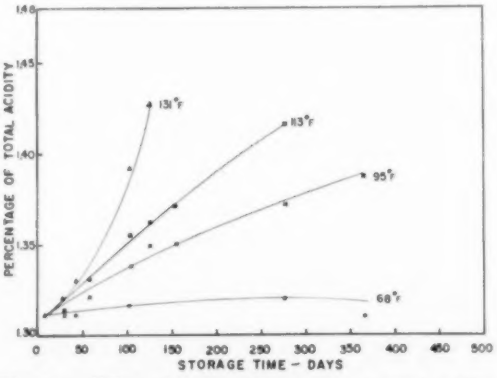

Figure 4. Effect of storage temperature on total acidity in canned tomato ketchup.

cate a more rapid increase in titratable acidity at higher temperatures. No significant increase in titratable acidity was found in the sample stored at 20° C (68° F) for a year. The rate of increase in acidity with time varies with storage temperature. The acid might be formed by degradation or oxidation of fructose in the tomato fruit. The remedy is to store the ketchup at low temperatures.

Lycopene and carotene. The lycopene content of the ketchup prepared for this experiment was 10.4 mg/100 g and the beta-carotene content 0.83 mg/100 g. It is evident that lycopene is the major carotenoid while beta-carotene exists only in small amounts. No significant change in these constituents was observed when the samples were stored at 55° C (131° F) for 19 days even though a definite darkening of the ketchup color was observed. Thus, the change in color of ketchup during storage was largely due to formation of brown pigments rather than chemical changes of the carotenoids.

When the sucrose was added to the ketchup during processing, the red color in the product suddenly became very intense. The reason for this phenomenon is not clear. It is possible that the distribution of lycopene which was concentrated in certain parts of the cell was changed by the addition of sucrose. Sucrose may have drawn more liquid from the cell to the outside fluid by osmosis, making the carotenoids more visible to the eye because of change in reflectivity.

Color. The color of the ketchup according to the U.S.D.A. color scale, remained at the "fancy" level when stored at 20° C (68° F), for 14 months. Higher storage temperatures caused a rapid change in color from bright red to brown. The canned ketchup changed from fancy to standard color grade in approximately 5 months at 35° C (95° F), 2 months at 45° C (113° F), and 1 month at 55° C (131° F). This indicates the importance of storing the ketchup at a low temperature range.

DISCUSSION

The rate of storage changes in tomato products may vary not only with temperature, but also with the type of product. The serum color of 26% tomato paste *(4)* darkened about twice as fast as that of ketchup, even though the total solids content of the paste (26%) was lower than that of the ketchup (32%). The slower rate of darkening in ketchup might be attributed to the fact that it is lower in tomato solids than the paste sample. Sugars in tomatoes consist mainly of glucose and fructose. Fructose is probably the constituent that causes browning reaction. Another possible explanation of browning differences is that pH affects the rate of darkening. The ketchup sample had a pH value of 3.7, while the tomato paste had a pH value of 4.2.

Ogle and Kramer *(7)* reported that the color of ketchup in bottle necks deteriorated very rapidly during the first few months of storage, while the color in the bulk of the product was maintained rather well and was not significantly altered until after 6 months' storage. During the final storage period (6–13 months) there was a significant loss of color. They believed that color deterioration during this period was related to the higher temperature of the summer months and indicated the need for additional work on controlled temperature storage. Results presented here show that storage temperature is important to ketchup stability.

Oxygen has been shown to be important to the black color formation in the neck of bottled tomato products *(3, 7)*. This was thought to be due to the oxidation of ferrous to ferric ions which then react with the tannin to form a black-colored product. With the introduction of air-tight bottle caps, deaeration of ketchup prior to filling, use of stainless steel equipment, control of head space, and selection of spices *(10)*, the black neck problem in bottled ketchup has been practically eliminated. In canned ketchup there is no leakage of oxygen into the can. However, browning of the serum and color degradation still proceeded when the storage temperature was high. This browning reaction is probably different from the oxidation reaction. The Maillard-type of reaction between carbonyl and amino groups, and the reaction between sugars and polyhydroxy carboxylic acids, the degradation of sugars to furfural, and the formation of hydroxymethyl furfural might partially explain the darkening of serum color. These chemical reactions were all accelerated by high storage temperatures. The extent of such storage changes increased with the length of storage time. Since ketchup samples with high serum color were invariably poor in flavor, every effort should be made to retard such changes by storing the product at 20° C (68° F) or lower.

The serum color test might be used as a quick objective criterion of ketchup quality. The test is easy to perform and is indicative of storage changes. The results presented here, however, show only trends of such chemical changes. More information is needed before the maximum serum color for good quality ketchup can be determined.

Other factors, such as preheating temperature and time, oxygen inclusion, contamination with iron or copper, concentration of tomato solids, and total solids in the ketchup, may also influence storage stability of canned ketchup. More work is needed in these areas.

SUMMARY

Canned tomato ketchup was made under controlled conditions and stored at temperatures varying from 20° C (68° F) to 55° C (131° F) for 14 months. The product was tested for chemical and color changes during storage. Results indicate that chemical changes occurred rapidly in ketchups stored at temperatures higher than 20° C (68° F). Serum color darkened and titratable acidity increased. High storage temperatures also contributed to decreases in amino nitrogen and degradation of color. The serum color test was found to be useful for detecting poor storage conditions in ketchup. To maintain good quality, storage at a temperature of 20° C (68° F) or lower is desirable. The importance of storage temperature on stability and chemical composition of ketchup is discussed.

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Influence of Ripeness Level on Organic Acids in Canned Tomato Juice

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Organic acids are important flavor constituents of tomatoes. Borntraeger *(2)* found that citric and malic acids were present in tomato juice. Hartmann and Hillig *(6, 7, 8)* reported that the concentration of these acids is 0.47% and 0.05%. Andrews and Viser *(1)* reported oxalic acid to be present at a concentration of 0.01% or less. Isherwood *(9)* and Marvel and Rands *(10)* presented a chromatographic method for isolating and determining organic acids. Bulen and Varner *(3, 13)* used a silicic acid column and an automatic constant-volume fraction collector for separating organic acids in plant tissues. They reported that the following organic acids are present in fresh tomatoes: acetic, formic, lactic, trans-aconitic, malic and citric.

More recently, Rice and Pederson *(11)* used the silicic acid column as described by Bulen *et al* for determining organic acids in tomato juice, but they modified the solvent schedule, the concentration of sulfuric acid in the aqueous phase, and the preparation of the sample. They reported that no previous purification of the sample is necessary for determining organic acids. Pyrrolidoncarboxylic acid was added to the list presented by Bulen *et al*. Since this acid was found only in processed tomato juice stored for two years, it is probably not present in fresh tomatoes.

In the present investigation, the silicic acid column chromatographic method was used to determine the effect of ripeness level on organic acids in canned tomato juice. Samples were freeze-dried prior to column chromatography and a solvent schedule similar to that suggested by Dame, Chichester, and Marsh *(5)* was used.

EXPERIMENTAL MATERIALS AND METHODS

Tomatoes. Improved Pearson variety tomatoes were planted in fine sandy loam soil in mid-April. The tomatoes were harvested in mid-October and sorted into 3 ripeness levels based on skin color. The Agtron E colorimeter was used for objective color measurement based on the ratio of green/red reflectance. The tomatoes were stored over-night at 32° F and made into juice the following day.

Canning procedure. About 150 lb of tomatoes for each lot were washed, stemmed, macerated in a Spike pump, and pumped to a wiped-film heat exchanger. Here, the product was heated at 230° F for 10 sec and then pumped into an American juice extractor with a 0.033 screen. The juice was mixed with 0.65% of sodium chloride, heated in a steam-jacketed kettle to 190° F, transferred into No. 2½ cans (⅜ in head space), and sealed while hot. Cans were heated in a rotary cooker at 212° F for 6 min, cooled in a water cooler to 110° F, and stored at 68° F until needed.

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Analyses. A representative sample of 40 tomatoes was taken from each ripeness level for chemical analyses and color evaluation.

Agtron E reading. The cross sectional area of the tomato halves was graded for color by the Agtron Model E colorimeter, which measured the ratio of green (546 mmc) to red (640 mmc) reflectance. This instrument is used by the State of California to supplement visual grading of canning tomatoes. The standard Agtron E readings for tomato color grades are as follows: well colored 0-46, fairly well colored 47-67, and off color 68 or higher.

Visual color. The visual skin color of the tomatoes was graded by a panel of seven persons. Each person scored the samples under a MacBeth standard light source. An excellent sample was scored 9–10, good 7–8, fair 5–6, poor 3–4, and very poor 1–2.

pH. The pH of the tomato slurry was measured with a Beckman glass electrode pH meter.

Titratable acidity. Titratable acidity was determined in a Beckman pH meter by titrating 10 g of the sample to pH 8.0 in 150 ml of distilled water with 0.1N NaOH.

Lycopene. The tomatoes were macerated in a Waring blender. Twenty-five grams of the macerate and 100 ml of 95% ethyl alcohol were mixed and filtered through a large sintered glass funnel of medium porosity. The residue was washed with small portions of a petroleum ether and alcohol mixture (2:1 v/v) until all yellow pigments were extracted. Alcohol was removed from the filtrate in a separatory funnel by adding 3% Na_2SO_4. The petroleum ether fraction was dried with anhydrous sulfate, and diluted to 100 ml. Ten milliliters of the petroleum ether extract was passed through a Hyflo-MgO (1:1) column and the lycopene was separated from the beta carotene by elution with 3% ethyl alcohol solution in petroleum ether. Optical densities of the fractions were determined for lycopene in a Beckman DU Spectrophotometer at 472 mmc.

Column chromatography of organic acids. Reagent grade silicic acid for chromatographic use was washed with distilled water and dried in an oven at 100° C for 24 hr *(3, 5)*. Twelve grams of prepared silicic acid was mixed with 8.2 ml of 0.5N sulfuric acid in a small beaker. The powder was slurried with a glass rod in 70 ml of chloroform and then transferred to the column. About 0.5 g of the lyophilized tomato powder was weighed and mixed thoroughly with 0.5 ml of 6N sulfuric acid. The mixture was combined with 1 g of dry silicic acid, and transferred quantitatively with a proper amount of chloroform to the column. Excess chloroform was drained through the silica gel column. Glass wool was placed above the sample to prevent disruption of the packed material while the solvents were being added. Solvents used to develop the column were: 100 ml of 5%, 135 ml of 15%, 100 ml of 25%, and 450 ml of 35% n-butyl alcohol in chloroform (v/v). Before use, the organic solvents were equilibrated in a separatory funnel with 0.5N sulfuric acid and then passed through double-layer filter paper to remove the small amounts of suspended water. The solvents were added successively to the top of the column through a reservoir. This permitted the application of compressed air which was freed from CO_2 by being passed through a 50% sodium hydroxide solution. Pressure was regulated by a clamp which maintained a flow rate of about 80 ml per hour. The effluent was passed through an automatic siphon device in 5.8 ml portions into test tubes and then transferred to Erlenmeyer flasks with a total of 20 ml of distilled water. Two drops of 1% phenol red indicator were added to the mixture. It was then rigorously agitated while being titrated with 0.01N sodium hydroxide until the end point was reached.

Paper chromatography. The non-volatile organic acids in canned tomato juice were investigated by paper chromatographic methods. The canned juice (Table 3, sample B) was freeze-dried to a powder under .08 mm Hg for 48 hr. Ten grams of the tomato powder was extracted with 100 ml of 95% ethyl alcohol, centrifuged, and the alcohol extract decanted. The residue was again extracted with 100 ml of 95% ethyl alcohol. The combined alcoholic extracts were evaporated on a steam bath to remove most of the alcohol. One hundred milliliters of distilled water was added, and the mixture filtered through a Whatman No. 1 filter paper. The filtrate was passed through a Duolite C-3 cation exchanger in the hydrogen form to remove metallic cations. After the column was washed with 100 ml of distilled water to recover all the free acids, the acids were adsorbed on a Duolite A-4 anion resin column in the hydroxide form. The column was washed with 500 ml of distilled water to remove the sugars and other constituents. The acids were recovered from this column with 100 ml of 2N NH_4OH, and the effluent was evaporated on a steam bath until most of the ammonia was expelled. The residue was diluted with distilled water to 50 ml, filtered through Whatman No. 1 filter paper, and then passed through a Duolite C-3 cation exchanger column in the hydrogen form. The column was washed with 100 ml of distilled water. The combined effluent was evaporated on a steam bath to a small volume, diluted with distilled water to 25 ml, and filtered. The filtrate was used for paper chromatography on Whatman No. 1 filter paper. Two developing solvent mixtures were used: (a) n-butanol-benzyl alcohol-water-90% formic acid, (50:50:10:1.1 v/v), and (b) phenol-water-90% formic acid (75:25:1 v/v) *(5)*. Chromatograms were developed for 20 hr at 70° F. The dried chromatograms were sprayed with a 0.04% solution of bromophenol blue in 95% ethanol at pH 5.8.

RESULTS

Color and chemical composition. The influence of ripeness level on the color and chemical composition of fresh tomatoes is shown in Table 1. As the tomatoes ripened, the average Agtron E readings decreased from 67 (Sample A, firm-ripe) to 34 (Sample C, soft-ripe), indicating an improvement in red color with ripening. When the tomatoes ripened, an increase in red reflectance and a decrease in green reflectance occurred. The visual color score of the tomatoes improved as ripening proceeded. Agtron E readings correlated well with the visual color grade and are reliable for grading color of fresh tomatoes. The pH of the tomatoes increased from 4.35 to 4.60 as they changed from the firm-ripe to the soft-ripe stage. The titratable acidity decreased with maturation. The lycopene content increased from 5.58 mg/100 g in firm-ripe tomatoes to 9.13 mg/100 mg soft-ripe ones.

Chromatography of organic acids. The organic acids in freeze-dried tomato juice samples were separated on a silicic acid column with n-butanol-chloroform mixtures. A typical sample for organic acids in a juice made from canning-ripe tomatoes is shown in Figure 1. Acetic, formic, pyrrolidonecarboxylic, malic and citric acids were present. Citric acid was most abundant, but pyrrolidonecarboxylic acid was present in a significant amount. The latter was reported to be formed from glutamic acid or glutamine by ring closure *(11)*.

TABLE 1

Influence of ripeness level on color and chemical composition of Pearson tomatoes

Sample	Ripeness level	Visual skin color score[1]	Color by Agtron E reading[2]	pH	Total titratable acid as citric, %	Brix @ 20° C	Lycopene, mg/100 g
A	Firm-ripe	5.7	67	4.35	0.412	4.83	5.58
B	Canning-ripe	8.5	47	4.43	0.383	5.02	7.87
C	Soft-ripe	9.2	34	4.60	0.330	5.32	9.13

[1] Visual skin color: Excellent 9–10, good 7–8, fair 5–6, poor 3–4, very poor 1–2.
[2] Color of tomato halves by Agtron E: Well-colored 0–46, fairly well-coored 47–67, off-color 68 or higher.

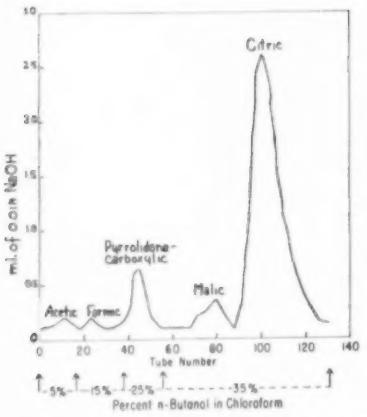

Figure 1. Silicic acid column chromatography of organic acids in tomato juice.

The non-volatile organic acids in canned tomato juice were investigated by paper chromatography of the various fractions from the silicic acid column. The organic acids were also identified without separation on a silicic acid column as described in the experimental section. The R_f values of the various organic acids are shown in Table 2. Citric acid was present in a larger quantity than malic or pyrrolidonecarboxylic acid. Lactic, succinic, and cis-aconitic acid were not found on the paper chromatograms.

Influence of ripeness level on organic acid content. The organic acids of tomato juices processed at various ripeness levels are shown in Table 3. Citric acid was a major organic acid in tomatoes, and was 73.2–80.1% of the total. As the tomatoes ripened, the citric acid decreased from 0.346% in the firm-ripe stage to 0.254% in the soft-ripe stage. There was a slight increase in malic acid from 0.0161% in firm-ripe tomatoes (Sample A) to 0.0239% in soft-ripe ones (Sample C). The pyrrolidonecarboxylic acid content varied from 0.0627–0.0835% and was 14.6–20.1% of the total organic acids. Acetic acid was present in a very small amount and this decreased slightly with maturation. The acetic acid may have been formed from de-esterification of the acetyl groups in the pectin molecule. Since the amount of this acid in tomato juice was only 1.0–1.1% of the total organic acid, it probably has no significant influence on the flavor of the juice.

TABLE 2

R_f values of non-volatile organic acids isolated from canned tomato juice

Peak No.	Solvent (a) n-butanol-benzyl alcohol-water, 90% formic acid (50:50:10:1.1 v/v)	Solvent (b) Phenol, 80 g, water, 20 ml; 90% formic acid, 1%	Organic acids
5	0.22	0.16	Citric
4	0.33	0.34	Malic
3	0.50	0.84	Pyrrolidonecarboxylic

DISCUSSION

The ripeness level of fresh tomatoes is important because it is related to the color, flavor, texture, and U.S.D.A. grade of the canned product. Results (Table 1) indicate the relationship between ripeness level, color, and composition of fresh tomatoes. The information gathered here shows a good correlation between the Agtron Model E reading and visual color grade. This instrument is effective and reliable for limiting the amount of poor-colored tomatoes in the canned product.

The pH value of the tomatoes increased with ripening. This explains at least in part why the tomatoes harvested during the 1958 season were higher in pH than usual. During the middle of that season, the weather conditions caused a large percentage of the tomatoes to ripen suddenly in a few days. The canners were not able to process the tomatoes delivered by the growers, and a limit on the maximum daily delivery was imposed. Since a larger amount of ripe tomatoes stayed on the vine for a longer time, the tomatoes delivered to the canneries from then on were overripe. This resulted in an increase in pH of the fresh tomatoes. Some canneries reported cases of pH values of 4.6 or even higher, and noted a greater percentage of spoiled canned whole tomatoes. Since sterilization of the heat-resistant microorganisms is greatly facilitated at pH values below 4.5, ripeness level could be an important factor influencing the pH of tomatoes and the conditions necessary for proper heat processing. Since titratable acids decrease with ripening, acidification of the tomatoes with citric acid to lower the pH below 4.5 would be desirable in preventing spoilage.

The decrease in titratable acidity of tomatoes was found to be due largely to a decrease in citric acid. Carañgal *et al (4)* indicated that immature tomatoes have a higher volatile acid and citric acid content. They reported that citric acid increases up to a certain point in the ripening stage and then decreases. The ripeness level of the fresh tomatoes probably determines the amount of citric acid.

The sum of the organic acids was slightly more than the titratable acidity (Tables 1 and 3). This might be explained by the fact that the method used here determines the sum of the free acid and the salt form of the organic acids present as ions. The titratable acidity which measured only the total free acids might therefore show a lower value than the sum of the free and combined acids.

Rice and Pederson *(11)* reported that 30% of the total organic acid in two-year old processed tomato

TABLE 3

Organic acids in tomato juices canned at different ripeness levels

Sample	Ripeness level	Agtron E reading of fresh tomatoes	Acetic			Formic			Pyrrolidonecarboxylic			Malic			Citric		
			Fresh basis	Dry basis	Total	Fresh basis	Dry basis	Total	Fresh basis	Dry basis	Total	Fresh basis	Dry basis	Total	Fresh basis	Dry basis	Total
			%	%	%	%	%	%	%	%	%	%	%	%	%	%	%
A	firm ripe	67	0.0048	0.0869	1.11	0.0018	0.0319	0.41	0.0627	1.14	14.6	0.0161	0.291	3.73	0.346	6.25	80.1
B	canning ripe	47	0.0045	0.0709	1.07	0.0014	0.0226	0.34	0.0835	1.32	20.1	0.0191	0.303	4.60	0.307	4.87	73.9
C	soft ripe	34	0.0039	0.0592	1.12	0.0013	0.0190	0.36	0.0640	0.97	18.4	0.0239	0.362	6.88	0.254	3.85	73.2

juice was pyrrolidonecarboxylic acid. In the present work, however, the pyrrolidonecarboxylic acid ranged from 14.6–20.1% of the total acid. This could be due to the difference in processing methods, storage conditions, and other variables. The work of Dame, Chichester and Marsh *(5)* on processed all-green asparagus supported the theory concerning pyrrolidonecarboxylic acid formation from glutamic acid or glutamine. The amount of heat received during processing or storage may determine the amount of pyrrolidonecarboxylic acid formed. Saravacos, Luh, and Leonard *(12)* found that glutamic acid is the major amino acid in tomato juice. This may explain the presence of pyrrolidonecarboxylic acid in canned tomato juice.

Results reported by various workers on organic acids in tomato juice and tomato products seem to conflict. These conflictions could be caused by variations in samples, analytical methods, storage conditions, or canning procedures. Hillig *(7, 8)* reported the presence of lactic acid in tomato juice, while Rice and Pederson *(11)* noted the presence of acetic, formic, pyrrolidonecarboxylic, malic, and citric acids. Results of the present investigation agreed with those of Rice and Pederson *(11)*, except that the quantity of pyrrolidonecarboxylic acid differed slightly.

SUMMARY

The influence of ripeness level on organic acids in Pearson tomatoes has been investigated by column chromatographic methods. The Agtron E was used to measure the color of the fresh tomatoes. The readings, based on the ratio of green/red reflectance, corresponded with the visual color grading and the lycopene content of the tomatoes. An increase in pH and a decrease in titratable acidity was found to occur during ripening. The importance of these factors in processing canned tomato products is discussed.

Citric acid was found to be a major organic acid in tomatoes, and was about 73–80% of the total organic acids. It decreased rapidly during ripening. Pyrrolidonecarboxylic acid was also an important organic acid in canned tomato juice. Acetic and formic acids were found only in very minute amounts. The importance of these acids to tomato juice quality is discussed.

Acknowledgment

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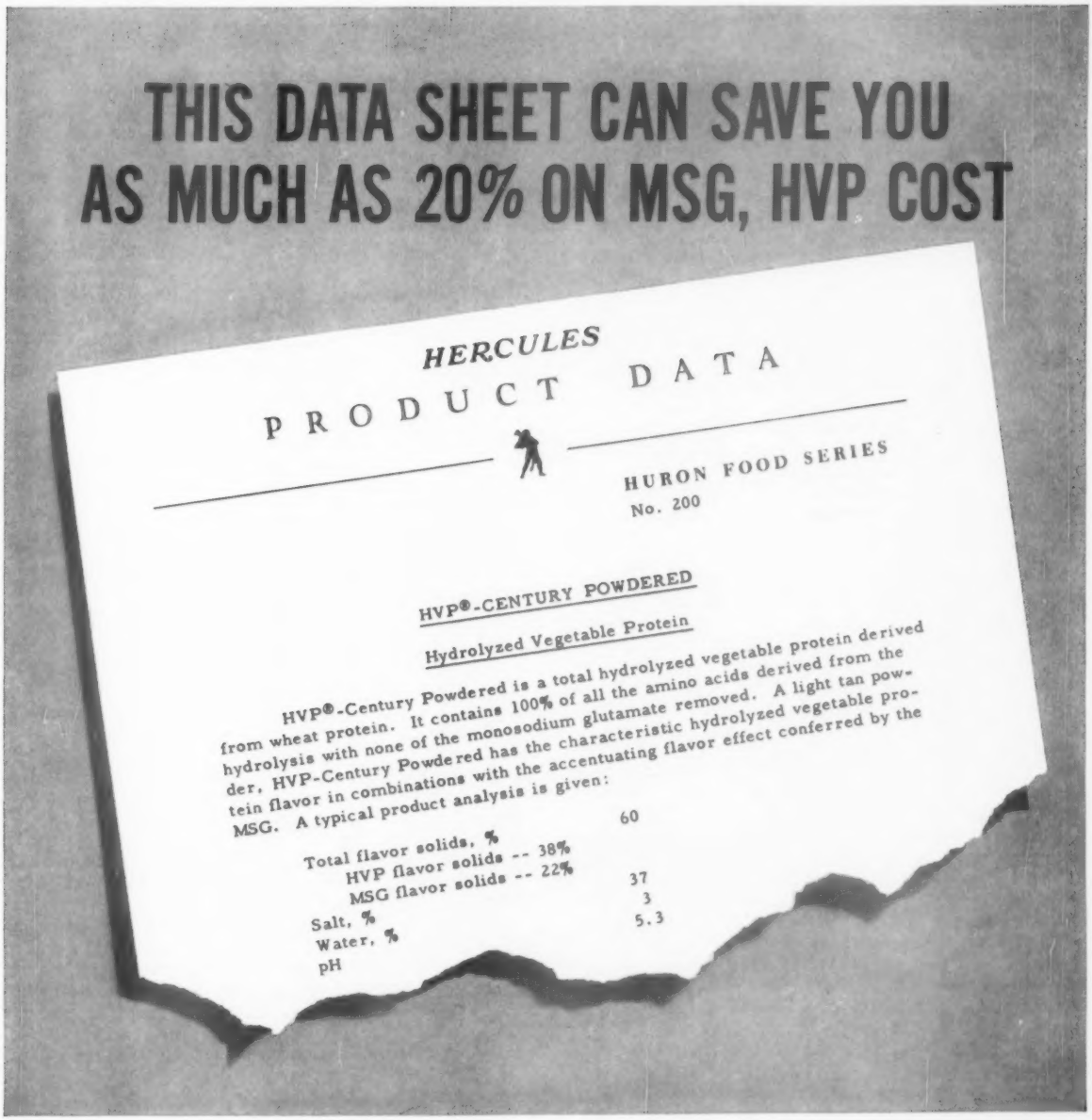
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Wine technology *Flor sherry*

Flor sherry production by submerged culture. C. S. Ough and M. A. Amerine.

The conventional method of flor sherry production, growing a film of flor yeast on the wine surface under aerobic conditions, requires five years or more for a desirable flor character to develop. Various trials with larger surface-to volume ratios have had such slight success that there has been little change in winery techniques. The flor character has not been chemically analyzed, but experienced tasters recognize the odor without difficulty. Aldehyde formation during fermentation, having some correlation with the intensity of the flor character, is a criterion for chemical measure of the flor flavor—a crude system at best. A more specific method of analysis is needed. The present studies were conducted to define more clearly such conditions of production as aeration, alcoholic content of the medium, and the effects on the yeast population of initially starting an aldehyde fermentation. In addition, a critical taste study was made to compare flor sherries aged in glass for one year and in small oak cooperage for nine months and for one year. The fermentation equipment used was tested by the sulfite method to show its capabilities for maintaining aerobic conditions with two types of paddles, by pumping-over with spraying through the headspace, and with increased pressure. The data show that with flat paddles nearly 100% of the oxygen would be available to the yeast with an input of 0.1 liter air per minute under the fermentative conditions described. Data on the alcohol content of aldehyde fermentations show that alcohol content should be kept below 15.0% by volume. Correlation between alcohol content and the maximum adehydes produced was −0.788, highy significant for the range studied.

Comparison of stirred versus pumped-over aldehyde fermentations show that the rate of aldehyde formation is about equal for either method, despite the fewer yeast cells in the pumped-over experiment. Investigation of the delay in starting an initial aerobic aldehyde fermentation indicates that the cause is a necessary acclimatization period, not establishment of a mutant yeast strain. Aging in oak cooperage (9 months or 1 year) produced flor sherries judged of a better quality than the same wines aged in glass. Data show some of the chemical differences between wines stored in oak and glass containers. (See page 155.)

Chemical inactivation *Enzymes in vegetables*

Chemical inactivation of enzymes in vegetables before dehydration. Rachel U. Makower.

Enzyme inactivation by chemical treatment at room temperature has been explored as a possible means of obtaining dehydrated vegetables of high quality. The initial experiments encouraged further investigations. The present paper describes methods developed and used for chemical enzyme inactivation and its testing. Nearly complete inactivation of tested enzymes was observed in shredded cabbage and pieces of apple, carrot, and celery which had been infiltrated under vacuum with a mixture of acid, alcohol, and a surfactant. None of these three components alone significantly inactivated the enzymes tested (catalase, peroxidase, acetyl esterase, phosphatase, and polyphenol oxidase) under the experimental conditions used. Enzyme inactivation increased with decreasing pH of the infiltration solution. Strong mineral acids with a pH of 1.5 or lower in the infiltration mixture were best enzyme inactivators. Anionic, cationic, and non-ionic surface-active compounds were tested as additives to the acid-ethanol mixture used for enzyme inactivation. A number of the surfactants synergistically enhanced the enzyme-inactivating action of the acid-ethanol mixture. The inactivation of enzymes increased with higher concentrations of the surface-active compound. In general, the anionic surface-active compounds were most effective synergistic enzyme inhibitors in the acid-ethanol mixture, while the cationic compounds were least effective. It is difficult to assess from the available data the relation between chemical structure of the surfactant and its effectiveness in the synergistic inhibition of plant enzymes. Of the enzymes tested, peroxidase appeared to be the most difficult to inactivate. (See page 160.)

Film packages *Pressure-volume relation during heat processing*

The pressure-volume relation in film packages during heat-processing. E. G. Davis, M. Karel, and B. E. Proctor.

This paper outlines investigations made on the internal pressure-volume relations in plastic containers, and includes:

a) Design of laboratory apparatus for the study of the pressure-volume relations.
b) Procedures used.
c) A record of observations made on film packages filled with either water or a food product during processing.

It is considered that the methods described may be suitable not only for use with heat-processed foods, but also for studying the pressure-volume relations in the "heat and eat" type of food packs. The expansion in volume with increases in internal pressure was studied on packages made from several films. Results showed that the expansion was sufficient to prevent high internal pressures developing during heat-processing. Internal pressures were shown to be related to the amount of material and enclosed air in packages of given dimensions when processed at a given temperature. Excellent agreement between calculated and estimated pressures was obtained on packages filled with water and processed at 212° F. The possible use of this technique as a guide for choosing the correct fill-in weights for packages to be heat-processed has been indicated. Differential pressures in film containers filled with water and several food products during processing at temperatures above 212° F under steam pressure were found to be relatively small except during the cooling operation. Cooling under superimposed air pressure was found to avoid the development of high pressures. The use of a package attached to a manometer as an indicator of the correct air pressure to be maintained during cooling has been described. (See page 165.)

Irradiation *Hams*

Control of oxidative changes in irradiated hams. Duane F. Hougham.

Irradiation of cured meats leads to certain oxidative changes which alter their flavor and color. Although the flavor changes are not particularly objectionable, it is desirable to control them as much as possible. This paper reports data from experiments planned with two objectives: first, to determine whether hams should be heated prior to or at some time interval after irradiation; second, to investigate the use of ascorbic acid and liquid smoke in protecting irradiated hams from oxidative changes. Although there is no apparent advantage in heating hams prior to or immediately after irradiation, it is apparent that no long interval of time should lapse between the two events. A smoke and ascorbate combination showed some antioxidant behavior. The smoke helped to mask the off-odors. The presence of ascorbate caused an unnecessary loss of nitrite and SH which led to an undesirable loss of pigment. (See page 170.)

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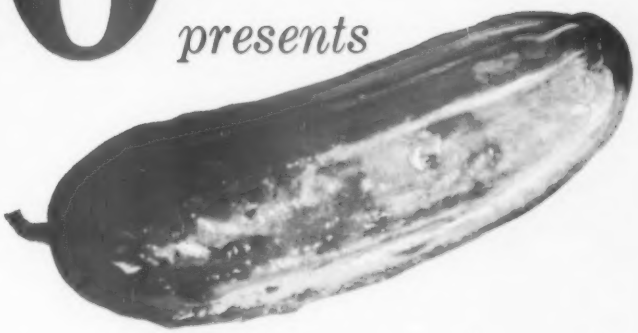

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Stability *Canned tomato ketchup*

Chemical and color changes in canned tomato ketchup. B. S. Luh.

One of the programs of the Department of Food Science and Technology at the University of California, Davis, concerns factors influencing storage stability of processed foods. Interest shown by canners in the stability of canned ketchup gave impetus to a study on the effect of storage temperature and time on chemical and color changes in canned tomato ketchup. Several objective tests for evaluating storage changes are presented. Canned tomato ketchup was made under controlled conditions and stored at temperatures varying from 20° C (68° F) to 55° C (131° F) for 14 months. The product was tested for chemical and color changes during storage. Results indicate that chemical changes occurred rapidly in ketchups stored at temperatures higher than 20° C (68° F). Serum color darkened and titratable acidity increased. High storage temperatures also contributed to decreases in amino nitrogen and degradation of color. The serum color test was found to be useful for detecting poor storage conditions in ketchup. To maintain good quality, storage at a temperature of 20° C (68° F) or lower is desirable. The importance of storage temperature on stability and chemical composition of ketchup is discussed. (See page 173.)

Ripeness level *Influence on organic acids in canned tomato juice*

Influence of ripeness level on organic acids in canned tomato juice. Fidel Villarreal, B. S. Luh, and S. J. Leonard.

In the present investigation, the silicic acid column chromatographic method was used to determine the effect of ripeness level on organic acids in canned tomato juice. Samples were freeze-dried prior to column chromatography. The Agtron E was used to measure the color of the fresh Pearson tomatoes. The readings, based on the ratio of green/red reflectance, corresponded with the visual color grading and the lycopene content of the tomatoes. An increase in pH and a decrease in titratable acidity was found to occur during ripening. The importance of these factors in processing canned tomato products is discussed. Citric acid was found to be a major organic acid in tomatoes, and was about 73–80% of the total organic acids. It decreased rapidly during ripening. Pyrrolidonecarboxylic acid was also an important organic acid in canned tomato juice. Acetic and formic acids were found only in very minute amounts. The importance of these acids to tomato juice quality is discussed. (See page 176.)

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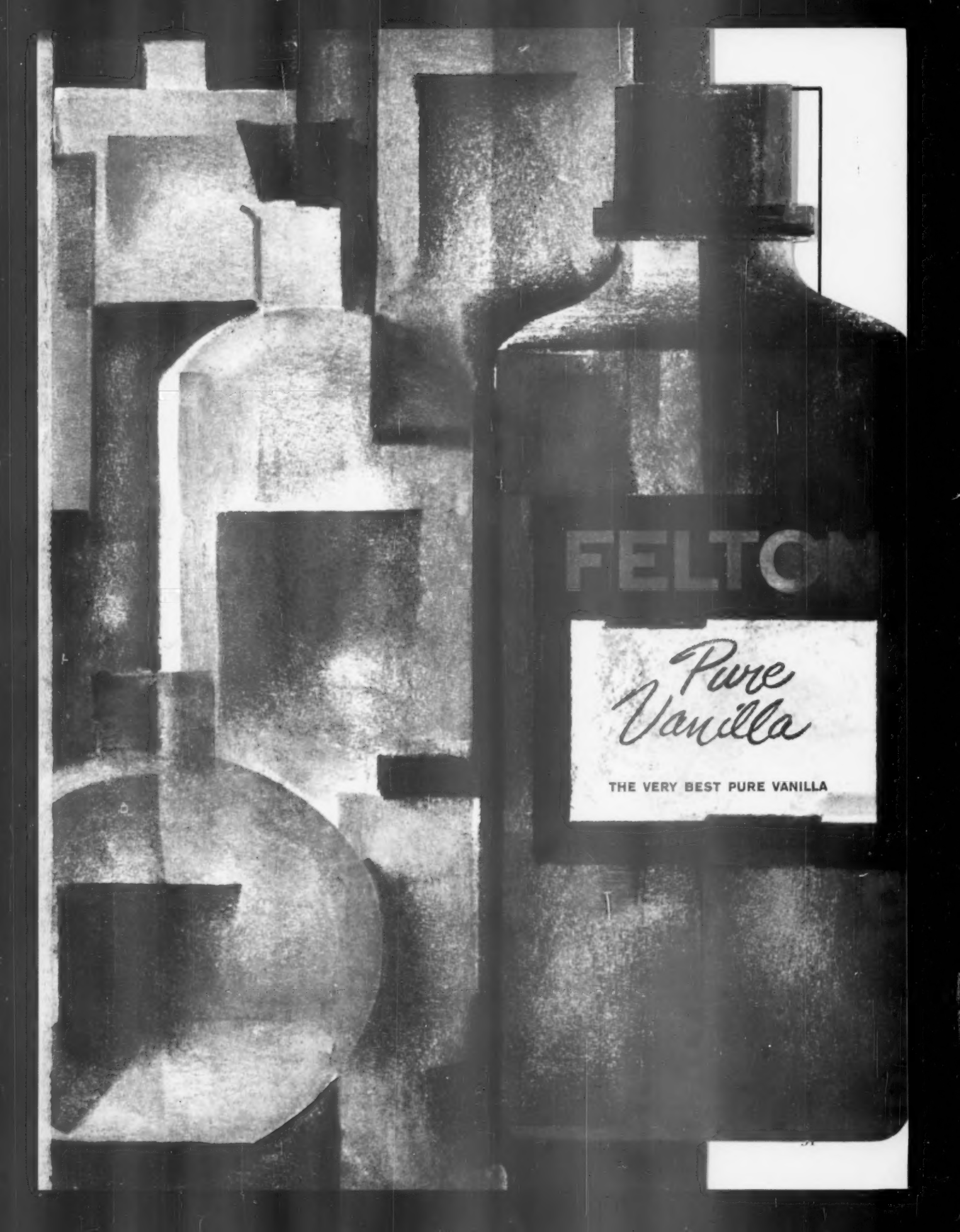
FUTURE MEETINGS FOR FOOD TECHNOLOGISTS

1960	
March 11-12	Eighth Annual Food Technology Short Course, "Impact of Convenience Foods," Memorial Student Union, University of Missouri. Sponsored jointly by the St. Louis and Kansas City Sections of IFT with the College of Agriculture, Columbia, Missouri
March 24-26	Symposium on Instrumental Methods for Analysis of Food Additives, Sponsored by the Food Technology Program, Michigan State University, National Canners Association, American Meat Institute Foundation, and the Packaging Institute, Kellogg Center, Michigan State University, East Lansing
April 4-6	51st Annual Meeting of the American Oil Chemists' Society, Baker Hotel, Dallas, Texas
April 12-13	Dairy Technology Conference: The New Food Additives Law and Antibiotics and Insecticide Residues; Product Judging; Procurement, Processing and Management Sessions. Sponsored by the Department of Food Technology and Divisions of the University Extension, University of Illinois, Urbana
April 19-21	Research and Development Associates 14th Annual Meeting, Congress Hotel, Chicago, Illinois
May 15-19	**Twentieth Annual Meeting of the Institute of Food Technologists, Fairmont Hotel and Masonic Temple, San Francisco, California**
May 19 to May 25 or 29	**IFT Pacific Rim Conference, Princess Kaiulani Hotel, Honolulu, Hawaii**
June 8-10	The 3rd Symposium on Gas Chromatography, Assembly Rooms, Edinburgh, Scotland
September 20-23	Pure Food Centenary, Royal Institution of Great Britain, London, England

1961	
May 7-11	**Twenty-first Annual Meeting of the Institute of Food Technologists, Hotel Statler, New York, New York**
1962	
June 10–14	**Twenty-second Annual Meeting of the Institute of Food Technologists, Fontainebleau Hotel, Eden Roc Hotel, Miami Beach, Florida**
1963	
May 19–23	**Twenty-third Annual Meeting of the Institute of Food Technologists, Pantlind Hotel, Civic Auditorium, Grand Rapids, Michigan**

An open invitation is extended to readers of FOOD TECHNOLOGY to send in to the Editorial Office, 11606 South Bell Avenue, Chicago 43, Illinois notices of annual or national meetings of interest to food technologists.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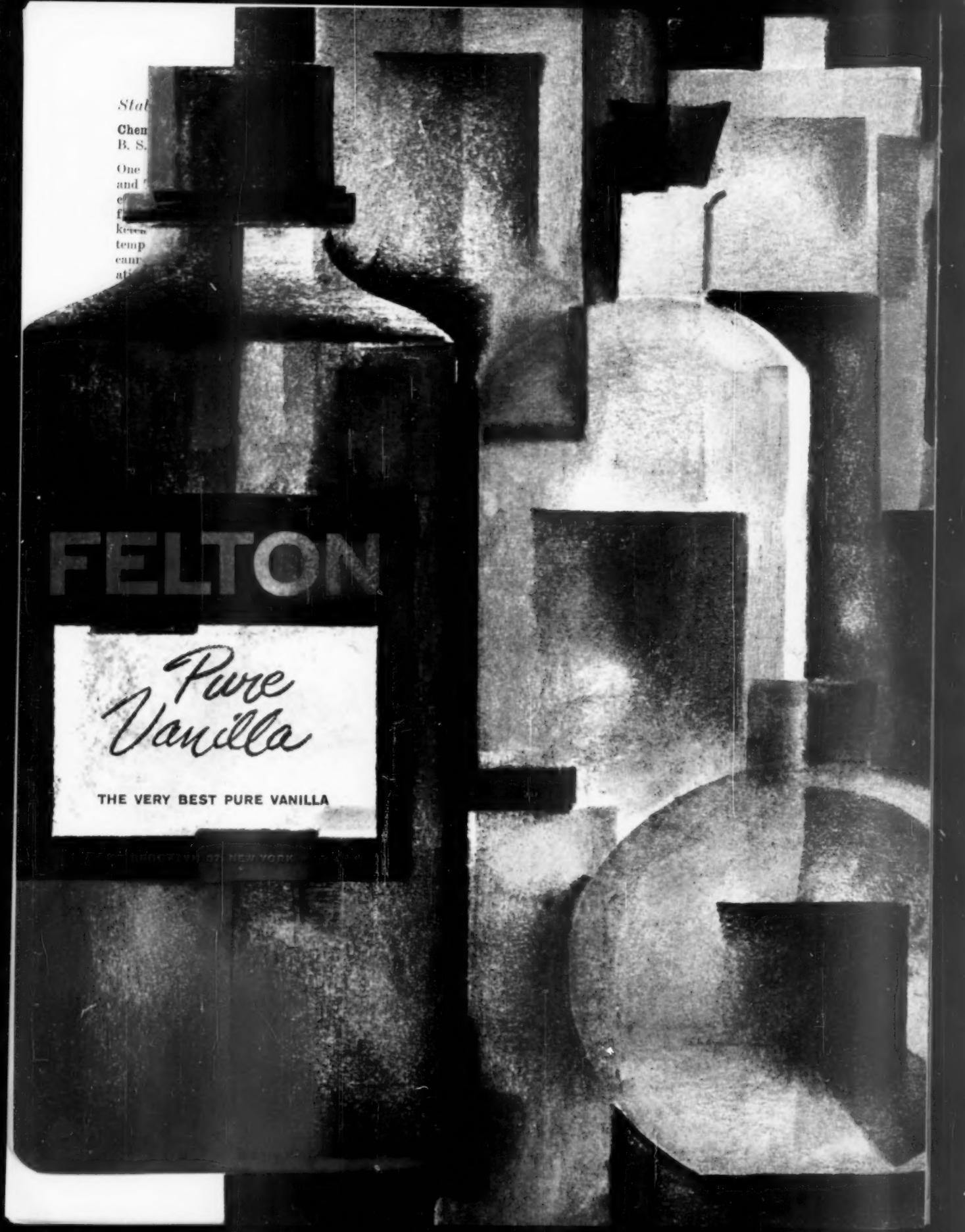
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Digest of Proceedings of Mid-Year Executive Committee Meeting (December 8, 1959)

A brief synopsis of the Mid-Year Meeting of the Executive Committee of the Council, December 8, 1959, is herewith placed on record.

Results of the Council Ballot on which recommendations made by the Executive Committee at the Mid-Year Meeting were listed, shows that 45 of 68 Councilors returned their ballots. All eighteen of the issues submitted to vote were approved by a significant majority.

New Journal Editors. Paul A. Buck, Beatrice Finkelstein, Willard B. Robinson, Kevin Shea, William Sulzbacher, and Joseph R. Wagner are the new Associate Editors of FOOD TECHNOLOGY, and M. A. Amerine, Bruce Morgan, C. F. Niven, and William Stahl are the approved editorial appointees for FOOD RESEARCH.

Publication of Pacific Rim Conference Papers. Since the Publications Committee, Program Committee, and Editor have no objection to separate publication, all papers to be presented at the Pacific Rim Food Conference will be released to the Program Chairman of the 20th Annual Meeting. The Program Chairman was authorized to arrange for publication of some or all of the papers, using his own discretion in determining which ones should be released.

Digest of Official IFT Business Meetings. In this issue and in future issues of FOOD TECHNOLOGY, brief digests of the proceedings of meetings of the Council and the Executive Committee of the Council will be published. Designated committee reports in digest form will be included. A minumum of space will be used in order not to infringe on the technical pages of the Journal.

Program Advisory Committee. A new Program Advisory Committee is established to make recommendations on long range policy in regard to Annual Meeting Programs. Recommendations shall include: (1) a list of the phases of food technology appropriate for inclusion in an Annual Meeting Program, (2) limitations on the number of papers and number of concurrent sessions appropriate for a given meeting. Members will be the current program chairman, incoming program chairman, and four members with previous program experience.

REPORTS

Treasurer's Report. Dollar-wise, the Treasurer, Dr. Arthur N. Prater, reports a gain from $193,523.17, October 31, 1959, to $218,769.82, December 7, 1959. Most of this increase is comprised of dues payments and advance exhibit fees for the Twentieth Annual Meeting at San Francisco.

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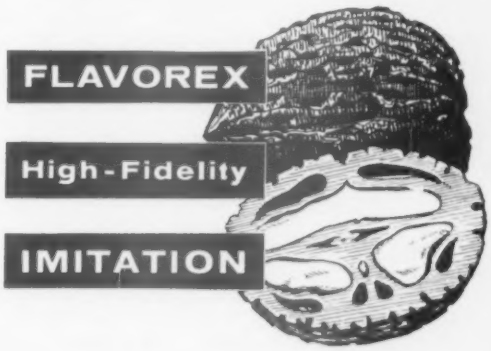

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A council resolution of May, 1958, authorizing a supervised agency account, was amended to the effect that the Executive Committee of the Council may authorize the deposit or withdrawal from the account of *"other available funds and the earnings thereon."* This is a technical correction and no change in the meaning or interpretation of the original resolution results.

Finance Committee and the Committee on Constitution and By-Laws. Two committees shall have a "continuing committee" status in the future: the Finance Committee, and the Committee on Constitution and By-Laws. For the former, the President will apoint three members (one annually) to serve a term of three years each, to augment the advisory resources of the Treasurer and Assistant Treasurer and otherwise assist them.

Two members shall be appointed annually by the President to the six-member Constitution Committee; they shall also serve for three years. The President shall designate one member as chairman of the latter Committee. In line with things constitutional, a thorough study of the Constitution will be made with the view of removing certain inconsistencies therein. It is anticipated that the Executive Committee and Council will initiate action on proposed changes at the Twentieth Annual Meeting in May.

Committee on Education. According to the Committee on Education, results of their July 1959 survey indicate that the demand for Food Technologists in the next five years with the approved type of training is more than double the supply of Food Technologists that were available during the past five years. In light of this, and of the fact that industry will need 1000 Food Technologists in the next five years, approval has been granted to revise and distribute our Career Brochure at a maximum expense of $6000. Revision is presently being implemented by the Committee.

Other Committees. There will be no formal function or facility set up by the Institute to translate Soviet or other foreign food research literature into English. The ad hoc committee on translation recommended that the Institute continue to keep its members informed through abstracts of overseas research in the food field, as it has done in the past, encouraging special contributions whenever a useful purpose can be served. Existing resources seem adequate for the present.

A complete report of the Ad Hoc Committee to Study Ways and Means of Advancing the Science of Food Technology will be presented at the May meeting.

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ITEMS RELATING TO ANNUAL MEETING OF IFT

The 1960 Annual Meeting at San Francisco will include installation of incoming officers at the Awards Banquet; the Business Meeting will be held immediately prior to the Appert Medallist's address. Following the Meeting, from May 20–25 will be the Pacific Rim Food Conference at which will be presented twenty to thirty papers, as well as numerous plant tours.

Future Annual Meetings are tentatively scheduled for New York, 1961, Miami Beach, 1962, Grand Rapids, 1963, and Washington, D. C., 1964. In view of this backlog and the fact that seven other Sections have submitted invitations to hold Annual Meetings, further invitations will not be considered until a later date.

OTHER ITEMS

Approval is effected for a new Health and Accident Insurance for Institute employees.

Public Relations in the form of news and award releases, annual meeting activities, and things of a similar nature will be handled at Institute Headquarters under supervision of the Executive Secretary.

In order to give appropriate recognition to the late Dr. Bernard E. Proctor, the Executive Committee moved that a commemorative resolution be prepared. The motion was seconded and unanimously approved.

IFT News

Hawaii for Food Technologists

Over thirty of your fellow food technologists (and some with their families) are already sniffing the Island Breezes. Join the following, who are either signed up for the Pacific Rim Food Conference, or are on the Committee for the event:

Dr. and Mrs. H. W. Adams; Dr. E. A. Beavens, Fruit & Vegetable Chem. Lab., USDA; Dr. C. D. Carpenter, Carpenter Associates; Mr. Frank B. Doyle; Dr. and Mrs. W. F. Geddes; Mr. W. L. Haley, Fisher Flouring Mills; Miss Dorothy L. Harrison; Doretta S. Hoffman; Dr. and Mrs. Imri J. Hutchings, H. J. Heinz Co.; Mr. and Mrs. David E. Lakritz, Florasynth Laboratories; Dr. H. Lineweaver, Western Regional Laboratory, USDA; Mr. and Mrs. Andrew E. Maren, Griffith Laboratories; Mr. and Mrs. Ernest Nordlinger, Roy D. Zeff Associates; Dr. J. W. Pence, Western Regional Lab., USDA; Mr. and Mrs. Carl D. Pratt, Atlas Powder Co.

Mr. and Mrs. Dan J. Ridenour; Prof. and Mrs. W. F. Robertson, Michigan State University; Mr. and Mrs. E. Rupp; Dr. and Mrs. Louis Sair, Griffith Laboratories; Dr. and Mrs. R. C. Sherwood, Sterwin Chemicals; Mr. R. S. Shumard, Hercules Powder Company; Dr. and Mrs. H. L. Sipple; Dr. G. F. Stewart, Univ. of California; Mr. W. B. Storey, Univ. of California; Mr. and Mrs. C. T. Townsend, National Canners Ass'n; Dr. Gladys Vail; Dr. R. H. Vaughn, Univ. of California; Mr. and Mrs. Ray B. Wakefield, Gerber Products Company; Mr. and Mrs. L. H. Walker, The Crescent; and Mr. and Mrs. R. Wilson.

Deadline for reservations, April 15.

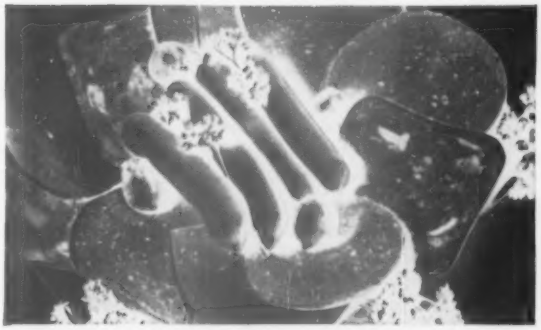
WASHINGTON, D. C.

The Washington, D. C. Section held seven meetings during 1959. One was a joint dinner meeting with the Maryland Section and one was the annual dinner meeting and ladies night. The other five meetings were held in the Auditorium of the National Canners Association. Each included a business and professional program followed by a social hour with refreshments. Outstanding speakers on a variety of technical subjects have appeared on the programs as follows:

February 3, 1959—The membership was privileged to have two speakers from the U. S. Department of Interior's Bureau of Marine Fisheries: Mr. H. L. Moore, Assistant Chief, Division of Biological Research, and Mr. Charles M. Butler, Coordinator of the Saltonstall-Kennedy Program of the Division of Industrial Research and Services.

March 20, 1959—Joint dinner meeting of the Maryland Section and the Washington Section was held at the Log Lodge, Agricultural Research Center, Beltsville, Maryland. The speaker for the evening was Dr. Aksel Olsen, National President of the Institute of

3 WAYS TO MAKE GOOD PRODUCTS BETTER



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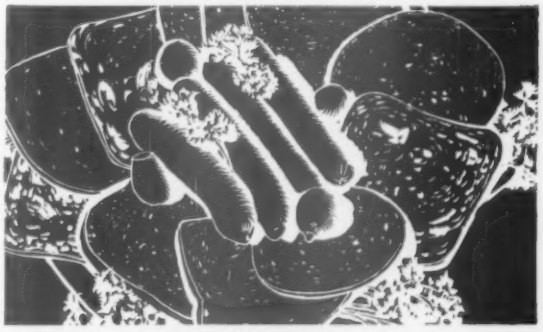

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Food Technologists in 1959, whose subject was "Industrial Research and Research Planning."

May 8, 1959—Dr. Oral L. Kline, Director, Division of Nutrition, Food and Drug Administration, U. S. Department of Health, Education, and Welfare, spoke on "Nutrition Activities in the Food and Drug Administration."

A second speaker, Miss Jean McNaughton, Regional Nutrition Officer, Food and Agriculture Organization of the United Nations, described "Current Developments in FAO Food Technology Programs."

June 30, 1959—Two speakers, Mr. Fred Hillig of the Division of Foods, Food and Drug Administration, U. S. Department of Health, Education, and Welfare, spoke on "Chemical Indices of Decomposition in Cod"; Mr. Cleve B. Denny of the National Canners Association, Washington, D. C. discussed "Preliminary Studies on the Heat Inactivation of Staphylococcus Enterotoxin."

September 29, 1959—The speaker for the evening meeting was Dr. Dale R. Lindsay, Assistant Chief, Division of Research Grants, National Institutes of Health, Department of Health, Education, and Welfare. He spoke on the subject of "Public Health Service Research Support in Food Science and Technology." Dr. Lindsay gave an interesting report on the administration of research grants and the N.I.H. program of research in the area of processing and quality, insecticide residues as food contaminants, food additives, and effects of processing on additives.

October 29, 1959—The program consisted of three firsthand research reports on the following subjects: "Preliminary Studies on the Isolation and Characterization of Meat Flavor Components," by Irwin Hornstein, Meat Laboratory, Eastern Utilization Research and Development Division, U. S. Department of Agriculture; "Bacteriological Survey of Precooked Frozen Foods," by L. R. Shelton, Food and Drug Administration, Department of Health, Education, and Welfare; "Recent Studies in Pectin Research," by Esam Ahmed, Department of Horticulture, University of Maryland.

December 4, 1959 (Annual dinner meeting and ladies night)—Mr. Ray B. Wakefield, President of the Institute of Food Technologists and Director, Research and Quality Control, Gerber Products Company, gave an interesting and informative talk on the subject "Babies are Important People."

Career Days—A committee on "Career Days" has been appointed to work with the University of Maryland on a program for 1960 to create interest in food technology among high school students.

Convention—Plans are being made by the Washington and Maryland Sections who will be co-sponsors of the 1964 Annual Meeting of the Institute of Food Technologists to be held in Washington, D. C.

The Washington Section had 150 members in December 1959 and hopes to increase its membership in 1960.

—*Elsie H. Dawson*

INDIANA

The March Meeting of the Indiana Section will be held Monday evening, March 21, 1960, at Turners Athenaeum, 401 East Michigan Street, Indianapolis, Indiana. The social hour begins at 5:30 p.m. E.S.T. and dinner will be served at 6:30 p.m. E.S.T. Dr. Bruce Morgan of the Continental Can Company will be the featured speaker for the evening.

The Indiana Section is encouraging all members to display their national membership certificates in their office or laboratory. Mr. Seb Davin is in charge of this project. We hope by this method to enhance the stature of the Institute of Food Technologists throughout the State of Indiana.

Section members worked on a display in the Memorial Center at Purdue University. The display showed the steps in can making and the different types of enamels. The cost of producing a single can of one product was broken down and shown in picture form. Another portion of the display indicated the education needed to be a Food Technologist. Special emphasis was given to the need for a good background in math, chemistry, physics, microbiology, and food processing. It is hoped that displays such as these will influence high school and beginning college students to consider the possibilities for education and employment in the food industry.

BRITISH COLUMBIA

A most interesting plant tour on January 12 was enjoyed by 45 members and their guests as they inspected the new Lake City Plant of Nabob Foods in suburban Vancouver.

Mr. John Walters, Director of Manufacturing, pointed out that this plant produced Nabob Tea, Coffee and Squirrel Brand peanut butter, although other products are warehoused and shipped from here. Mr. Walters conducted the tour through the single story-straight line flow factory and warehouse.

Many examples of new techniques were observed. Pneumatic conveyors, wet dust collection systems, automatic electronic batch weighing equipment, automatic tea bag machines, etc., contributed to the efficiency of this well laid out modern plant.

The company kindly provided coffee and refreshments in the cafeteria, and Mr. Walters answered further questions. E. Watson thanked Mr. Walters and his company on behalf of the group.

CHICAGO

At the January Meeting of the Section, Mr. James W. Bannon of Booz, Allen and Hamilton, Management Consultants, spoke on new product development. Mr. Bannon went about describing the organization, planning and development of new products in a very methodical fashion, segregating various components

of this complicated process into the area of responsibility and decisions needed from management, marketing, research and sales. The first step in any new product development program must be made by top management, which has the responsibility of defining the area of interests of the company and consequently the kind of products which could be successfully marketed by the company. Mr. Bannon maintained that failure to do this can cause a grevious waste of talent and effort. To illustrate his point, he showed that when independent inventors develop ideas and submit them to management, very few of these ideas ever bear fruit, largely because the outside inventor is unacquainted with the product needs and desires of the company. Need for critical decision making at a very early stage of any product development program was pointed out with great emphasis. These decisions should concern profit potential, ability of the company to make the product and merchandise it, as well as availability of technical and sales personnel. If these facts are not examined early in the history of any new product idea, product development work is sometimes undertaken without the company having the ability to either make or sell the item. Mr. Bannon also pointed out various steps needed to conduct successful acquisition or merger operations, as well as time and cost factors involved at various stages of the development of an idea into a product.

S. CALIFORNIA

Neither flu, smog, nor Virus M could hold down the attendance of the Southern California IFT'ers as they met at Roger Young auditorium January 20, 1960.

The gavel was turned over by out-going chairman Henry Espoy to in-coming chairman Gordon Miller, who in turn presented the out-going chairman with a scroll for his outstanding year during 1959. After the treasurer's report and a resume of meetings to come (featuring National IFT President-Elect I. J. Hutchings at the March 16th meeting and later on in the year an all day meeting in Long Beach). Henry Espoy, formerly of Barnett Laboratories in Long Beach and presently with Terminal Testing Laboratories, Inc., delivered the feature talk of the evening on the subject "Color In Your Foods."

Mr. Espoy traced the progress in color in foods from its inception to present day conditions. The complete procedure of certification of coal-tar colors was covered as was the story of the delistment of Orange No. 1, Orange No. 2, and Red No. 32. Mr. Espoy explained how D&C No. 14 had been used temporarily and that now Citrus Red No. 2 was approved for use by the citrus industry in some sections of the country until Sepetmber 1961. It was further stated by Mr. Espoy that the cut off date on the usage of Yellow No. 1, Yellow No. 2, Yellow No. 4, and Yellow No. 3 had been extended till April 6, 1960. The Color Additive Amendment was discussed as were recent trends and new innovations in natural colors such as Carotene and Annatto. The group was informed by Mr. Espoy that future Carotene would be made from raw materials containing from two to three times as much color and that manufacturing procedures would make a new and superior type of Carotene available.

Chairman Gordon Miller announced that the following members had agreed to serve as officers during 1960: Vice Chairman—Dr. Cora Miller, Department of Home Economics, UCLA; Secretary—Bob McGrath, Chas. Pfizer Company; Treasurer—Clive Ross, Firmenich Company.

—D. K. Wright

FOOD ADDITIVE NEWS

The **1959 Joint National Conference of Food and Drug Administration and the Food Law Institute, Inc.** was held in Washington, November 16 and 17, 1959. More than seven hundred people attended the two-day meeting at which important problems under the Food Additive Amendment and Federal Food, Drug, and Cosmetic Act were discussed by representatives of FDA, industry, and consumer groups. The complete proceedings of the meeting are being published in the *Food, Drug, Cosmetic Law Journal*, starting with the December, 1959, issue.

The annual meeting of the **Food Protection Committee, National Research Council,** and the Liaison Panel was held in Washington, December 9 and 10, 1959. A feature of the meeting was a symposium on "Special Problems in the Toxicology of Food Additives" under the chairmanship of Dr. J. M. Coon, Jefferson Medical College. Dr. J. M. Barnes, Director, Toxicology Research Unit, Medical Research Council of Great Britain was the speaker at the annual dinner.

The **Food Protection Committee** has prepared two new reports, "Problems in the Evaluation of Carcinogenic Hazard from the Use of Food Additives," and "Principles and Procedures for Evaluating the Safety of Food Additives." Printed copies of both reports will be available shortly from the National Research Council, 2101 Constitution Avenue, Washington, D. C. Two active projects of the Food Technology Subcommittee include the revision of "The Use of Chemical Additives in Food Processing" (the so-called "Green Book") and the preparation of a statement on functions and benefits of food additives.

New officers for the **Industry Committee of the Industrial Liaison Panel** include L. H. Burt, Chairman, Hercules Powder Co., Wilmington 99, Delaware; E. F. Binkerd, Vice-Chairman, Armour & Company, Chicago 9, Illinois; R. L. Hall, Secretary, McCormick & Co., Inc., Baltimore 2, Maryland.

Literature

BOOKS

CONVERSION FACTORS AND TECHNICAL DATA FOR THE FOOD INDUSTRY. Compiled by C. G. Harrel and R. J. Thelen. 6th Edition. Burgess Publishing Company. Minneapolis. 1959. 1426 pages.

This book contains a compilation of a vast amount of data, and conversion factors from a wide range of subjects. As might be expected from the technical backgrounds of the compilers, much of the information presented deals specifically with cereal and baked products. To the best of my knowledge, this is the first time so much useful data, specific to the food industry, has been compiled into one book.

The data are presented in 14 major subject areas. These include: conversion factors — measurements and equivalents; foods and food ingredients; general bakery data; plant and plant equipment, nutrition; temperatures. Much of this information will be quite familiar to the reader. However, there is a great deal of interesting subject matter set forth in an easy-to-read and clear manner. In any book of this size, the subject index is of importance. In this book a fairly complete, well-detailed index is given. Without it, the voluminous data would be useless.

It would be presumptuous of me, in a short review such as this, to attempt a detailed discussion of the subject matter but certain topics are worthy of note. There is an excellent chapter on analytical and physical data on foods, containing about 150 pages of reference tables in food composition. The section on nutrition discusses the nutrients in foods, and should help to answer the many questions arising in this most important aspect of our business.

The chapter on temperatures, among other things, contains an interesting listing of "important temperatures to remember." Beyond this each reader will find, depending upon his interest, other subject areas which summarize data useful to his field.

As mentioned previously, this book is weighted in the area of cereal and baked products. Besides this, some of the information presented may soon become, or perhaps already has become obsolete. The section on Federal Standards of Identity is presented in a limited way. Since this is a constantly changing area, this material perhaps should be left to other publications which can devote more space to a more complete coverage.

This book is recommended as a reference book for all persons in the food and allied industries whose positions require them to have available a ready source of information on technical or scientific subjects. Although all of the information will not always be of value, there is enough to suit each individual to make the purchase price worthwhile. I have already found the answers to a number of problems in my copy.

CHARLES FELDBERG
Brooklyn, New York

LITERATURE ON FOOD RESEARCH AND TECHNOLOGY[a]

Commodities

Das Getreide. Matei Rohrlich and Bernhard Brückner. Berlin, Verlag A. W. Hayn's Erben. 1959. 2 vols. 394 p., 90 illus., 2 color pl., 121 tables. DM. 50.50. General survey of technology, current researches and their applications for cereal grains.

Milk and milk products in human nutrition. Stanislaw K. Kon. Rome, Food and Agriculture Organization of the United Nations.[b] (FAO Nutritional studies no. 17). 1959. 25 p. paper, $0.75.

Olive oil. Rome, Food and Agriculture Organization of the United Nations.[b] (Commodity reports; fats and oils no. 8). July 1959. 34 p. paper, $0.25. Review and prospects of production and consumption.

Report of the fourth session of the FAO group on grains to the committee on commodity problems. Rome, Food and Agriculture Organization of the United Nations.[b] 1959. 32 p. paper, $0.25. Statistics, trends and international agreements.

Rice. Food and Agriculture Organization of the United Nations.[b] (Commodity reports no. 10). August 1959. 67 p. paper, $0.25. Review of, and trends in, rice production, export, etc.

Zucker und Zuckerwaren. Albrecht Rincke. Berlin, Verlag A. W. Hayn's Erben. 1959. 140 p. DM. 19.50.

General

Australian and South African canned deciduous fruit industries. Frank C. Elliott. U. S. Department of Agriculture, Foreign Agricultural Service publication M61. 1959. paper, 24 p. Apply to Service, Washington 25, D. C.

Automating the Manufacturing Process. George F. Hawley. N. Y., Reinhold. 1959. 147 p. $4.95.

Chemistry as a profession. J. L. Riebsomer. Cambridge, Mass., Bellman Publishing Co. (Vocational and professional monographs no. 104). 1959. 20 p. paper, $1.00. Another in the series of guides to vocations.

Evaluation of protein nutrition. Washington, Committee on amino acids, National Research Council, publication 711. 1959. 61 p. paper, $2.00.

Factors influencing the palatability, vitamin content, and yield of cooked beef. U. S. Department of Agriculture, Home Economics Research report no. 9. October 1959. 83 p. paper, $0.50 (from Superintendent of Documents, Washington 25, D. C.).

Food for survival after disaster. Raymond C. Hutchinson. N. Y., Cambridge University press. 1959. 90 p. $2.75. Food for survival after airplane accidents, ship wrecks, etc.

Outlines of Enzyme Chemistry. 2nd ed. J. B. Neilands and Paul K. Stumpf. N. Y., Wiley. 1959. 411 p. $8.50.

Review of food consumption surveys. Rome, Food and Agriculture Organization of the United Nations.[b] 1958. 107 p., ca. 100 tables. Breakdown of budget amounts, percentages, by types of food for many areas of the world, based on published sources.

The Sequestration of Metals. Robert L. Smith, B.Sc., Ph.D. F.R.I.C. N. Y., The Macmillan Company. 1959. 256 pages. $8.50. After defining and explaining the nature of sequestration, the chelate ring, etc., industrial applications including those in the agriculture, food, beverage, pharmaceutical industries are considered.

Statistical forecasting for inventory control. Robert G. Brown, N. Y., McGraw-Hill. 1959. 232 p. $7.75. Principles of forecasting maximum reasonable demand.

[a] Material for this list should be sent to Walter L. Necker, Assistant Librarian, Quartermaster Food and Container Institute for the Armed Forces, 1819 W. Pershing Road, Chicago 9, Illinois.

[b] Available in the U.S.A. from Columbia University Press, International Documents Service, 2960 Broadway, New York 27.

ABSTRACTS

General Foods Research Center
Selected by H. A. Campbell

ANALYTICAL METHODS

Paper chromatographic investigations of cereal extracts for substances of the tannin group.

TIEMANN, F. AND BLUMENBERG, F. W. *Arztl. Forsch. 13*, I/193–I/195 (1959).

Paper chromatographic investigations of wheat, rye, barley, oats, and maize indicated that only barley contains the tannin-like substances gallocatechin and epicatechin. Wheat, rye, and barley contained mixts. of flavone-flavonole corresponding to chrysin-galangin or apigenin camphor oil. Chlorogenic acid could be identified in all cereals.

Nuclear magnetic resonance spectra of alkaloids. I. The complete structure of lunacrine and lunine.

GOODWIN, S., SHOOLERY, J. N. AND JOHNSON, L. F. (Natl. Heart Inst., Bethesda, Md.). *J. Am. Chem. Soc. 81*, 3065–9 (1959).

The structure was proposed for lunacrine, the principal leaf alkaloid of *Lunasia amara* Blanco, on the basis of chemical, u.v., and nuclear magnetic resonance studies. The complete proof of structure by means of nuclear magnetic resonance spectroscopy of lunacrine and the companion alkaloid lunine is presented.

The effect of Tween 80 on pH indicators.

SARGENT, J. D. AND WALKER, J. R. L. (Massey Agr. Coll., New Zealand). *Lab. Practice 8*, 343 (1959).

Tween 80 in concns. of 0.1% and 0.2% causes changes in the color of bromthymol indicator giving false pH readings at pH's of about 7.

BIOLOGICAL SCIENCES

Biochemistry

The enzymic decomposition of cellulose.

HALLIWELL, G. (Rowett Research Inst., Aberdeen, Scotland). *Nutrition Abstr. and Revs. 29*, 747–59 (1959).

The present state of knowledge of the degradation of cellulose by cellulolytic organisms is reviewed with regard to mechanisms, rumen microorganisms, estn. of the amt. of cellulose decomposed, and types of cellulases.

Diet and the action of cortisone on protein metabolism.

GOODLAD, G. A. AND MUNRO, H. N. (Univ. of Glasgow, Scotland). *Biochem. J. 73*, 343–8 (1959).

The effect of cortisone on nitrogen balance and on liver protein and ribonucleic acid content has been studied on rats receiving diets varying in protein and in energy content. The catabolic action of cortisone on the body as a whole was independent of the protein and energy content of the diet, whereas the anabolic action of cortisone on the liver was greater when the dietary energy intake was high. It has been concluded that loss of protein from the carcass after cortisone administration results in a greater supply of amino acids for other organs and in consequence protein synthesis in these organs and urea formation are increased.

Sulfhydryl-disulfide interchange.

JENSEN, E. V. *Science 130*, 1319–23 (1959).

It has recently become evident that disulfide rearrangements take place in the absence of added reducing and oxidizing agents and appear to be chain-type reactions initiated by very small amts. of SH compds. This article summarizes reported examples of sulfhydryl-disulfide interchange phenomena in an attempt to expain aspects of protein denaturation, blood clotting and mitosis.

A lipoperoxidase factor in soya extracts.

BLAIN, J. A. AND STYLES, E. C. (Royal Coll. of Sci. and Technol., Glasgow, Scotland). *Nature 183*, 1141 (1959).

The oxidation of linoleate by cytochrome c and by lipoperoxidase were compared in order to det. whether exts. of soybean and other plant materials owe some of their unsatd. fat oxidase activity to the presence of hematins as well as to lipoperoxidase. It was concluded that cytochrome c bleaches carotene by the destruction of preformed peroxide rather than by the coupled concurrent oxidation of linoleate, whereas the lipoperoxidase causes increase in conjugated diene in the presence of β-carotene. Hematins and lipoperoxidase were most active at pH of 3.8 and 5 to 6, resp., showing a different response to H ion concn. A difference in activity between buffer exts. and water exts. of soybeans was also noted.

Linoleic and linolenic acids: Their oxidation by normal and diabetic rats.

LOSSOW, W. J. AND CHAIKOFF, I. L. (Univ. of California, Berkeley). *Proc. Soc. Exptl. Biol. Med. 101*, 857–60 (1959).

Oxidation of linoleic and linolenic acids, both randomly labeled with C^{14}, to CO_2 was compared in fasted and glucose-fed normal rats; normal and diabetic fed rats; and insulin-treated and untreated diabetic fed rats. Administration of glucose spared oxidation of both linoleic and linolenic acids. Oxidation of linoleic acid by ad lib.-fed diabetic rat exceeded that by similarly fed normal rat. Insulin spared oxidation of polyunsatd. fatty acids in the diabetic rat.

Biosynthesis of fatty acids and cholesterol considered as chemical processes.

CORNFORTH, J. W. (Natl. Inst. for Med. Research, London, Engl.). *J. Lipid Research 1*, 3–28 (1959).

A survey from the viewpoint of an organic chemist, of the processes leading to biosynthesis of fatty acids and of cholesterol. 120 references.

Microbiology

Studies on the bacteriological quality of frozen meat pies. IV. Longevity studies on the coliform bacteria and enterococci at low temperatures.

KERELUK, K. AND GUNDERSON, M. F. (Campbell Soup Co., Camden, N. J.). *Appl. Microbiol. 7*, 327–8 (1959).

Sterile chicken gravy samples inoculated with equal numbers of *Escherichia coli*, *Aerobacter aerogenes*, *Streptococcus faecalis* and *Streptococcus faecalis* var. *liquefaciens* were stored at −6° F. The coliforms decreased whereas the enterococci remained relatively const. *Aerobacter aerogenes* and *S. faecalis* var. *liquefaciens* were more resistant to the lower temp. storage than were *E. coli* and *S. faecalis*.

Ethylene oxide sterilization of spores in hygroscopic environments.

OPFELL, J. B., HOHMANN, J. P. AND LATHAM, A. B. (Cutter Labs., Berkeley, Calif.). *J. Am. Pharm. Assoc. 48*, 617–19 (1959).

Bacillus globigii spores were exposed to ethylene oxide after being dried on several types of surfaces. The viability tests, after exposure, showed significant effects were produced by the presence of the hygroscopic substances, glycerin, and filter paper. Absence of hygroscopic substances appeared to increase the resistance of *B. globigii* spores to gaseous ethylene oxide sterilization.

Nutrition

A comparison of several methods for estimating the nutritive value of proteins.

RIPPON, W. P. (Australasian Food Research Labs., New South Wales). *Brit. J. Nutrition 13*, 243–60 (1959).

Protein quality has been estd. from (1) nitrogen-balance methods, (2) change in body-wt. and protein efficiency ratios, (3) changes in carcass N, (4) changes in liver N, (5) analysis of the protein for amino acids, and (6) changes in the creatinine:N ratio in urine. Measurement of N balance by the balance-sheet

method seemed the most reliable provided (1) N intake was maintained at the same level for all tests and (2) the metabolic fecal and urinary N of the test animals were detd. before each test. In addn. to biol. value, this method also makes available other useful values such as net protein utilization and true digestibility.

Evaluation of protein in foods. IV. A simplified chemical score.

McLAUGHLAN, J. M., ET AL. (Dept. of Natl. Health and Welfare, Ottawa, Can.). *Can. J. Biochem. Physiol. 37*, 1293–9 (1959).

It is shown that a simplified chem. score, based upon the lysine, methionine, and cystine content of foods, correlates well with results of P.E.R. detns. on the same foodstuffs, and hence, may be useful as a simple screening procedure for the evaluation of protein in foods.

The experimental production of arteriosclerosis: Response of the avian artery to intramural cholesterol and other insoluble substances.

NETSKY, M. G., CLARKSON, T. B. AND STOKES, D. (Wake Forest Coll., Winston-Salem, N. C.). *Am. J. Pathol. 35*, 1081–9 (1959).

The injection of cholesterol into the adventitia of the avian artery caused a reaction consisting of foreign-body giant cells, cholesterol slits, and increased amts. of connective tissue. Free fat was abundant in the tissue spaces and within macrophages at the site of injection. The intramural injection of paraffin or $BaSO_4$ induced a response at the site of injection which differed from that caused by cholesterol. The response was largely of lymphocytes when paraffin was injected, and of connective tissue with $BaSO_4$.

Effect of a high-fat diet on growth and blood sugar of the rat.

FETTER, D. AND NEIBLE, E. A. (Brooklyn Coll., N. Y.). *Metabolism: Clin. and Exptl. 8*, 762–8 (1959).

Young female rats on a high-fat diet grow better and have blood sugar levels about 20 mg.% higher than those fed a low-fat, high-carbohydrate diet. After adrenalectomy the blood sugar levels in the rats fed high-fat diets become more like the normal values recorded for the rats on a high-carbohydrate diet. It is suggested that a high-fat diet may stimulate secretion of anterior pituitary hormones and thereby indirectly cause increased production of adrenal cortical hormones, which could induce a high blood sugar.

Rice intolerance in infants: masked food allergy?

JOHNSTONE, D. E. *Ann. Allergy 17*, 250 (1959).

Infants with a positive family history of allergy show a high susceptibility to rice intolerance and to subsequent development of major respiratory allergies. Symptoms manifested include colic, diarrhea, rhinorrhea, cough, wheezing and increased hunger.

The effects of a soybean product on thyroid function in humans.

VAN WYK, J. J., ET AL. (Univ. of North Carolina, Durham). *Pediatrics 24*, 752–60 (1959).

Studies relating to the development of goiter and hypothyroidism in an infant fed a soybean milk-substitute product suggest that a goitrogenic agent was present in the product causing increased iodine requirement due to impaired thyroid hormone synthesis. Investigations indicate that different lots of soy vary widely in this respect and that heat treatment lessens, but does not abolish, the goitrogenic effects.

Improvement in nutritional value of peas by cooking.

KIENHOLZ, E. W., JENSEN, L. S. AND MCGINNIS, J. (Washington State Univ., Pullman). *Proc. Soc. Exptl. Biol. Med. 102*, 35–8 (1959).

Growth and feed utilization of chicks and poults fed diets contg. high levels of peas (*P. sativum*) were markedly improved by cooking or autoclaving peas prior to mixing into diets. Greatly increasing caloric content of diets by adding tallow did not give a growth response comparable to that obtained by simply cooking the peas. Alkali-insol. fractions of peas depressed chick growth but alkali-sol. fractions did not. It is suggested that peas contain a growth inhibitor for birds that is inactivated by heating treatments employed.

Iron. I. Absorption and metabolism. A review of recent contributions.

WOODRUFF, C. W. (Vanderbilt Univ., Nashville, Tenn.). *Borden's Rev. of Nutrition Research 20*, 61–73 (1959).

Several studies have shown that the amt. of medicinal iron absorbed varies inversely with the bulk of the meal with which it is fed. Studies have also indicated that children show a relatively higher retention than adults, and that there is little difference in iron absorption in normal and anemic children. Except in unusual cases of hypochromic anemia in infancy and early childhood there is little likelihood that the adminis-

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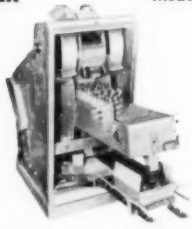
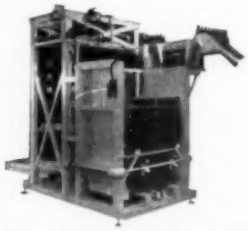


This high-speed unit handles a wide range of can, jar and bottle sizes (packed in single tier cases)—at speeds up to 30 cases per minute. No changeover necessary to switch from cans to bottles of same dimension.

PALLET LOADER AND PALLET UNLOADER



Straight-stack all standard cases and bottle sizes without mechanical adjustment, at speeds up to 30 cases per min. Designed for 36″ x 36″, 36″ x 48″, and 36″ x 60″ standard pallets. Can be furnished for other sizes. Available individually, or in combination.

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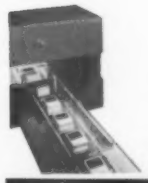

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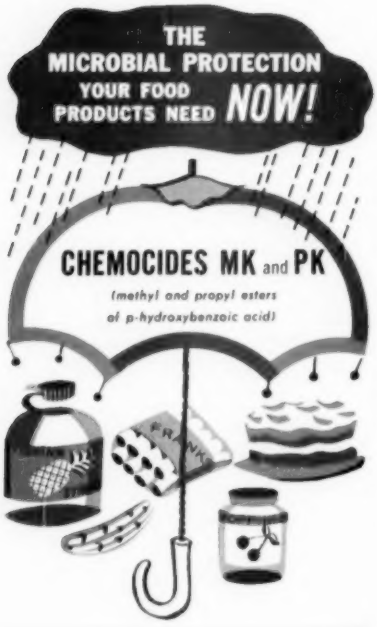

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tration of copper will increase the effectiveness of iron supplements. The claim that cobalt improves the utilization of iron is doubtful. There is no evidence that the various organic iron examples which have been introduced in the last few years are more effective than ferrous sulfate.

Toxicology

Methyl alcohol in fermented beverages.

ANON. *Assoc. of Food and Drug Offic. Quart. Bull. 23*, 171–3 (1959).

In the production of alcoholic beverages, the fermentation process produces some methyl alc. The author reviews the toxicology and pharmacology of methyl alc. and briefly summarizes the mechanisms of its formation during fermentation. He points out that ethyl alc. (obviously present in alcoholic beverages) has an inhibitory action on the oxidations of methyl alc. Thus, the small amts. of methyl alc. present in fermented beverages constitute no health hazard.

Study of the toxicity of dust with use of the in vitro dehydrogenase technique.

MARKS, J. AND NAGELSCHMIDT, G. (Tuberculosis Reference Lab., Cardiff, Wales). *A. M. A. Arch. Ind. Health 20*, 383–9 (1959).

Macrophages were incubated with dust and their capacity to reduce tetrazolium was measured. The amt. of dust which reduced the activities of a standard cell culture by 50% was called the toxic dose. Good correspondence was found between the latter method and observations on fibrogenic activity of the same or similar dusts made in intact animals.

Toxicity of amine-extracted soybean meal.

GREENBERG, J., ET AL. *J. Agr. Food Chem. 7*, 573–6 (1959).

Residues of soybean meal, after extn. with primary, secondary, or tertiary org. amines, were toxic for the chick when incorporated, at levels of 20 to 40%, in an otherwise adequate diet. Triethylamine-extd. cottonseed meal, black-eyed peas, soybean protein, zein, and Cerelose were toxic in varying degrees. Triethylamine-extd. gelatin and casein were well tolerated.

FOOD AND FOOD TECHNOLOGY

Antioxidants

Relative antioxidant activities of the seven tocopherols.

LEA, C. H. AND WARD, R. J. (Low Temp. Research Sta., Cambridge, Engl.). *J. Sci. Food Agr. 10*, 537–48 (1959).

The antioxidant activities of the 7 tocopherols and of the unmethylated parent compd. (tocol) have been compared in several systems. The 3 tocopherols unsubstituted in the 5-position (δ, γ and η) were more effective than the other 4 in protecting carotene in mineral oil soln. at 75°.

See multi benefits in new malts.

OLSON, W. J. *Food Eng. 31*, 103–4 (Oct. 1959).

Recent findings indicate that malt flour has a food preserving effect (imparts oxidative stability) beyond that attributable to its antioxidant properties. Fats thus treated are not rancid although they reach peroxidase levels at which they would be expected to be rancid.

Cereals and Grains

Lysine and threonine supplementation of rice.

ROSENBERG, H. R., CULIK, R. AND ECKERT, R. E. (E. I. du Pont de Nemours and Co., Inc., Newark, Del.). *J. Nutrition 69*, 217–28 (1959).

When proper balance has been reached between the first two limiting amino acids further improvement in protein quality should be attainable if both amino acids are added simultaneously and in the proportion of the body's requirements. It should be possible to add this amino acid mixt. in graded amts. and to obtain increasing growth responses until a third amino acid, or other nutrient, becomes limiting. This concept has to the authors' knowledge not been tested experimentally. A study was undertaken, therefore, to check this theory using rice as the test protein and the growth of the male weanling rat as the biol. criterion. The results of this study, reported here, support in general the above concept for supplementation of proteins with the first two limiting essential amino acids.

Nuts

Nutmeg poisoning.

GREEN, R. C. *J. Am. Med. Assoc. 171*, 1342–4 (1959).

The toxic factor of nutmeg is known to be myristicin, a constituent of the volatile oil of nutmeg. Nutmeg in doses of 5 g. or more produces a marked depressive action on the central nervous system, as well as a less prominent stimulating effect. The clinical course may be severe, with coma, shock, and acidosis as its main features.

Taste Testing

Odor difference test with applications to consumer preferences.

MRAK, V., ET AL. *Food Research 24*, 574–8 (1959).

Two expts. on detecting odor differences between unknowns of stated strengths and a standard are presented. In the first case the standard was presented just before the unknown and the tester was asked to respond weaker or stronger. In the second expt. the standard was given immediately before a

series of 3 unknowns and again the tester's responses were weaker or stronger. These expts. indicate great variation between individuals in their ability to detect odor differences and in their response to the different ways of presenting the samples.

Gelatin

Configuration transitions in gelatins in non-aqueous solution.

VEIS, A. AND ANESEY, J. (Armour and Co., Chicago, Ill.). *J. Phys. Chem. 63*, 1720–5 (1959).

The behavior of gelatin in formic acid (FA)-dimethylformamide (DMF) and in ethylene dichloride-dichloroacetic acid has been examd. The studies cover (1) an analysis of the optical rotatory power and rotatory dispersion as a function of solvent compn.; (2) the configurational changes as described by viscosity measurements; and (3) the roles of aggregation and degradation as detd. by light scattering. The light scattering data indicate that no aggregates are formed in the FA-DMF mixts. These data are consistent with the assumption that a hydrogen-bonded helical structure is formed when FA/DMF < 1 but this helix is not the same as that of native collagen.

Marketing

The "ordinary" $125-billion market.

ANON. *Fortune 60*, 132–5 (Sept. 1959).

The amts. spent by Americans for food and clothing for the last 30 years are reviewed and future prospects are considered. In 1959, Americans will spend close to $70 billion for food and this will continue to increase as population and wages increase. However, the percentage of income spent on food may decrease from the present 37% to 35% by 1970, because of the natural limit on how much can be consumed as well as the decline in food prices.

Pet food industry 'on the march' meeting indicates.

ROGERS, D. E. *Feedstuffs 31*, 1; 133 (Sept. 12, 1959).

Pets owned by U. S. families add up as follows: dogs 23.8, cats 25.5 and birds 12.2 million. Marketing studies indicate that 90% of dog owners buy canned food in the course of a year and 80% buy dry dog food; however, dry food continues to become more important. A large potential market is said to exist for premium-priced canned cat foods.

Milk and Milk Products

Effect of hard water on the heat stability of skimmilk powder.

SARGENT, J. S., BIGGS, D. A. AND IRVINE, D. M. (Ontario Agr. Coll., Guelph, Can.) *J. Dairy Sci. 42*, 1800–5 (1959).

Protein stability in reconstituted skimmilk is adversely affected by high Ca and Mg levels in the reconstituting water. The heat stability, as measured by the Storrs stability test, is a function of total hardness, and is not changed by varying the Ca to Mg ratio. The effect of Mg ions can be balanced by sequestration with equivalent quantities of Na hexametaphosphate, but with Ca more than equivalent quantities of this sequestering agent are needed. Sequestration of a portion of the hardness of very hard waters is suggested as a possible method for avoiding premature protein coagulation during heat processes.

ORGANIC CHEMISTRY

Clathrates.

MANDELCORN, L. (Westinghouse Research Labs., Pittsburgh, Penna.). *Chem. Revs. 59*, 827–39 (1959).

This review summarizes the literature pertaining to the synthesis, classification, structure, properties, and applications of clathrates. The problems encountered in the detn. of the thermodynamics of

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clathrate systems and in the analysis of thermal decompn. data are discussed.

PACKAGING

Research, born of necessity, is bringing progress in self extinguishing plastics.

ANON. *Modern Plastics 37*, 90–8 (Nov. 1959).

Self extinguishing materials are those which will not burn once an outside source of flame is removed. Self-extinguishing plastics available today are listed along with manufacturer's name and ASTM testing data. The chart is broken down into resins and molding compds., film, sheeting, and laminates, and plastic foams.

Epoxidized polyolefin resins.

GREENSPAN, F. P., JOHNSTON, C. AND REICH, M. *Modern Plastics 37*, 142; 144; 146; 226 (Oct. 1959).

A new family of epoxy resins, contg. epoxy groups in both internal and terminal portions, is described with particular attention to the curing characteristics and mechanical properties. It is expected that these resins will open up new applications of epoxy resins in castings, laminates, adhesives, and coatings.

PHYSICAL METHODS

Determination of molecular dimensions from rheological data.

PETERLIN, A. (Univ. of Ljubljana, Yugoslavia). *Die Makromolekulare Chemie 34*, 89–119 (1959).

A survey of the existing possibilities of molecular size and shape detn. from rheological expts. is given. Porous sphere models, necklace models, packed coils, corpuscular and linear particles are among the configurations considered.

RADIOCHEMICAL TECHNOLOGY

$C^{14}O_2$ respiration from labeled compounds in feeding-trained rats.

KIRK, M. R., LEPKOVSKY, S. AND TOLBERT, B. M. (Univ. of California, Berkeley). *Metabolism: Clin. and Exptl. 8*, 769–76 (1959).

Rats were trained over a period of several months to eat their daily food in 2 hrs. The effect of feeding on the C^{14} respiration patterns of radioactive acetate, glucose, fructose or glycine injected 1, 8.5 and 22 hrs. after feeding was studied, and cumulative breath excretion as well as the specific radioactivity of the $C^{14}O_2$ was measured. Feeding increases total CO_2 production and total glucose, fructose and glycine oxidation to CO_2. The initial oxidation rate of these compds. is also sharply increased. The concept of the specific dynamic action of food is applied to most of the findings.

Industry

IN MEMORIAM

Dr. W. E. Pyke, head of the Colorado State University chemistry department, died on December 22, 1959, at Houston, Texas. His death was attributed to a coronary occlusion. He was 63.

Dr. Pyke, on the Colorado State University staff since 1924, began work as an assistant instructor in chemistry. He was made head of the department in 1945. He also served as secretary of the Colorado State University Research Foundation.

Dr. Pyke received his A.B. degree from Baker University at Baldwin, Kansas, in 1918 and his Ph.D. from the University of California, Berkeley, in 1940. He was a veteran of World War I, having served in the Ordnance Corps, Quartermaster Corps and Chemical Warfare Service of the U. S. Army. He also served as camp supply officer at Camp Kendrick, New Jersey.

He was a member of a number of professional societies, including the American Chemical Society, Sigma Xi, Institute of Food Technologists (charter member), American Society of Experimental Biology and Medicine, American Association of University Professors, American Society for Advancement of Science, Swiss Chemical Society, and Colorado-Wyoming Academy of Science. Dr. Pyke was author of a large number of scientific publications.

Survivors, in addition to Mrs. Pyke, include three daughters—Mrs. Leonard C. Wilson and Mrs. Lela Geisendorfer, both of Fort Collins, and Mrs. James Randall of Mineral, California—a sister, Miss E. Marie Pyke, and a brother, Cecil Pyke.

PERSONNEL

Mr. George S. Congdon has recently been appointed Production Manager of the Orange-CRUSH Company of Evanston, Illinois, according to Louis Collins, vice-president and general manager of the company. Prior to joining Orange-CRUSH, he has had wide experience with major companies in chemical, technical service and managerial research capacities.

William C. Lytle has retired as vice-president of Atlas Powder Company after a diversified career spanning almost all of the company's history. A native of Leadville, Colorado, Mr. Lytle joined Atlas as a chemist shortly after receiving his bachelor's degree from the University of Colorado in 1917. He served in many capacities until he was elected vice-president in June, 1958, with key responsibilities for the Explosives Divisions and assigned corporate staff activities.

The appointment of **Dr. Benjamin Borenstein** as a food technologist in Nopco Chemical Company's Fine Chemicals Division has been announced by Dr. Harold Yacowitz, Director of Applied Research for the division. He will be engaged in the development of products and processes as related to the food industry. Dr. Borenstein was previously employed by Fearn Foods, Inc., Franklin Park, Illinois, as a research chemist. He is a graduate of Rutgers University with a B.S. degree in chemistry and M.S. and Ph.D. degrees in food technology.

Polak's Frutal Works, Inc. (P.F.W.) held its annual sales meeting on December 14–16 at their plant in Middletown, New York. Pictured are all participants. Standing left to right: E. Johnson, D. Moncino, P. Delaney, L. Pais, E. Sturman, J. Donnelly, K. Freiberg, A. Krol, A. J. de John, Dr. J. S. Jellinek, IFT Member **M. Preiser,** F. Marshall, H. Lonergan, E. Buday, J. Melody; seated left to right: E. Vles, Dr. R. Boucher, A. H. Micheels, E. Polak, W. R. Lloyd (Ass't Managing Director of Soflor, Ltd., England), B. Polak (Pres.), J. Meihuizen (Co-Director of P.F.W., Holland), J. ter Veer, L. Kirsh, H. Roger (Far Eastern representative).

Dr. Bernard S. Schweigert will head the College of Agriculture's new department of food science at Michigan State University. Dr. Schweigert is now director of research and education for the American Meat Institute Foundation, Chicago.

B. S. Schweigert

The Board of Trustees of MSU approved the framework for the new department several months ago. It combines the work involved in the processing of agricultural products into a single department. Some staff members working with food processing in the departments of horticulture, animal husbandry, dairy husbandry, poultry science, farm crops and agricultural engineering will be regrouped into the new department.

Dr. Schweigert, author or co-author of more than 150 scientific publications in the field of biochemistry, nutrition and meats research, will join the staff effective May 1, 1960. He earned his B.S., M.S. and Ph.D. degrees from the University of Wisconsin in biochemistry and physiology from 1943 to 1946 and did teaching and research at Wisconsin and at Texas A. and M. College before joining the American Meat Institute Foundation in 1948. He has been jointly associated with the University of Chicago while at the Meat Institute Foundation.

M. J. Thomas D. P. Langlois

Dr. M. J. Thomas has recently been named director of chemical market development in the Market Development Division of the A. E. Staley Manufacturing Co. He was born in Yazoo City, Mississippi, has a 1936 bachelor's degree from Mississippi State College, and master's and doctor's degrees, in 1938 and 1941, from Vanderbilt University. In June 1942, he joined the company as a research chemist, moved to Market Development as an assistant in 1948 and was named assistant manager of that division in 1953.

Also promoted is **Dr. David P. Langlois.** who now occupies the position of director of administrative services for Market Development. Formerly he was a technical information scientist in the Research Division.

John R. Miller has recently been promoted Eastern Sales Manager of Florasynth Laboratories, Inc., New York City. Since 1956 he served as a flavor specialist assigned to Florasynth's midwest sales office. During World War II, Miller was assigned to the Quartermaster Corps, Subsistence Depot, Sidney, Australia, with responsibilities for developing sources of food supply in Australia, and in 1952 he was assigned as officer-in-charge, Munich (Germany) Procurement Office where he helped establish a processed meat program to American standards. He also assisted in eliminating tuberculosis in Austrian dairy cattle.

J. R. Miller

The board of directors of General Foods Corporation recently elected **Dr. Robert N. DuPuis** vice president—research. Dr. DuPuis will asume leadership of General Foods' corporate research activities, including the supervision of the research center at Tarrytown, New York. He will, however, remain on the board of directors of Philip Morris, Inc., where he has been vice president in charge of research and development since 1952. Dr. DuPuis earned his Ph.D. in organic chemistry at New York University in 1934, after graduating from the University of Illinois with Phi Beta Kappa honors and an A.B. degree in 1931. He held managerial research positions with S. C. Johnson & Son, Inc., at Racine, Wisconsin, from 1947 to 1952 and with Miner Laboratories at Chicago from 1935 to 1947.

R. N. DuPuis

Ac'cent International, manufacturers of the food flavor enhancer, Ac'cent, has announced the appointment of **Dr. William H. Bartholomew** as manager, microbiological development. A bio-engineer, he comes to Ac'cent International from the Pabst Laboratories Division of Pabst Brewing Company, where he was assistant director of research. Dr. Bartholomew, who received his Ph.D. at Pennsylvania State University in 1941, will conduct liaison work between research, development, and production departments looking toward the design and operation of an industrial fermentation plant, according to G. B. Hamilton, vice president of Ac'cent International, a division of International Minerals & Chemical Corporation. IMC announced in mid-summer the development of a new fermentation process to produce pure monosodium glutamate.

W. H. Bartholomew

Recently appointed as sales manager and assistant sales manager of the Dewey and Almy Chemical Division, W. R. Grace & Co., are **Richard W. Haward** of LaGrange, Illinois, and **Robert J. Douglass** of Marblehead, Mass.

R. W. Haward R. J. Douglass

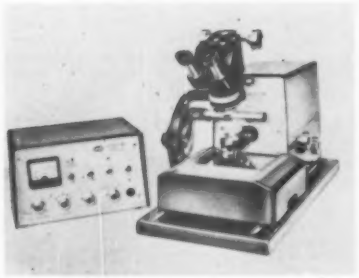
Haward received an A.B. degree from Harvard College in 1949 and joined Dewey and Almy in 1953. Prior to his promotion, he served as Midwest sales manager for can, closure and industrial compounds.

Douglass began with Dewey and Almy in 1956 serving as container products sales engineer and product manager for side seam cements. He received an A.B. from Cornell University in 1948 and an M.B.A. from Cornell's Graduate School of Business and Public Administration in 1949.

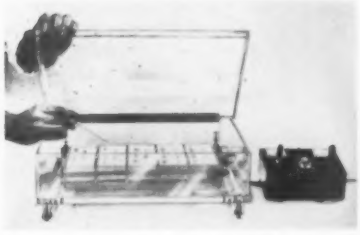
Paul G. Miller has been named research director of Adolph's Food Products Manufacturing Company, Burbank, California. A graduate of the University of Wisconsin (Ph.D., Biochemistry), Dr. Miller was formerly Assistant Research Director of the Thomas J. Lipton Co., Hoboken, N. J.

P. G. Miller

EQUIPMENT

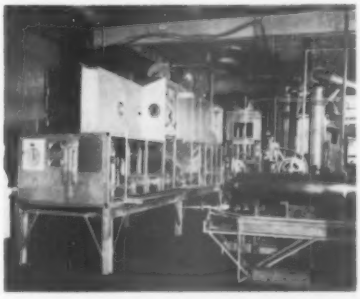


Now available to electron microscopists is the LKB 4800 ultrotome Ultra-Thin Sectioning Microtome for routine production of ultrathin sections — single and serial. Sections up to 145 microns can also be cut in sequence with the ultrathin. Two independent specimen feed systems are used, thermal and mechanical. Lateral and rotary motions of the specimen mechanism have been eliminated. Engineering principles introduced are preselected and automatic fall rate of specimen arm past knife edge (five speeds), elastic knife retraction, arc-shaped specimen and knife holders, and a dual illumination system. Literature and further information are available from LKB Instruments, Inc., 5451 Randolph Road, Rockville, Maryland.



Development of a multiple-cell paper electrophoresis apparatus, designed for running one to five samples simultaneously at five different pH values from 3.3 to 9.3, with special organic buffers said to yield complete separations in one hour has been announced by Microchemical Specialties Company, Berkeley, Calif. Misco's apparatus with organic buffers and special reference dyes identifies fractions of microgram to milligram quantities of acids, bases, carbohydrates, amino acids, proteins, plant hormones, alkaloids, drugs, and other compounds. Known samples can also be separated for quantitative analysis. For more information, write Microchemical Specialties Company, 1825 Eastshore Highway, Berkeley, California.

Submergible pumps available in 200 to 514 gph are now available from the Thomal Beckett & Co. Dallas, Texas. Pumps are brass, and said to be designed to operate while fully or partially submerged in lubricant fluids, water, citrus juices, milk coolers, and other liquids. Retail price is from $20.00 to $30.00.



The Henszey Company, Inc., has announced completion of a pilot plant facility at its Watertown, Wisconsin plant. This equipment will be used for their own development projects, and will be available to industry in general as a research and development aid. It includes a new pilot model spray dryer with horizontal air suspension, and various types of collectors, all interchangeable, said to insure maximum recovery of various types of products. The plant is equipped with a fully automatic control panel, and new evaporating and heating components have been erected to allow a full testing program for most types of materials.

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WANTED: FOOD TECHNOLOGIST & WINE CHEMIST to locate in central California with large National concern. Experience with food or beverage desired. Excellent opportunities and company benefits. REPLY BOX 749, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

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FOOD TECHNOLOGIST of 8 to 10 years industrial experience to assume position of senior responsibility in Quality Control and Development for multi-plant food processing operations involving canning, freezing and dehydration. REPLY: Curtice Brothers Co., 328 Main St. East, Rochester 4, N.Y.

AVAILABLE: Food Technologist, B.S. Purdue 1948. Some graduate work. Experience—canning, carbonated beverages and dairy products. Desire Chicago location. REPLY BOX 762, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

AVAILABLE: FOOD TECHNOLOGIST, 10 yrs. experience dairy, canned and frozen foods. Desires situation in Product Control, Product Development or Technical Service. REPLY BOX 759, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

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WANTED: FLAVOR CHEMIST—Leading New York Essential Oil and Flavor Company expanding operations. Wonderful opportunity for advancement. Salary open. Write giving details with confidence since replies handled by company head. Our employees are aware of this position. REPLY BOX 761, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

TECHNICAL SALES

Food Technologist to service processors of foods, bottlers, ice cream and candy manufacturers. Excellent opportunity with progressive flavor manufacturer. Write age, education, previous experience, and other pertinent data. REPLY BOX 763, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

POSITION WANTED: Experienced Technologist desires position in charge of Quality Control and/or New Products Development. Doctorate degree in dairy manufacturing. Extensive dairy and food products background. WRITE BOX 751, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

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FOOD TECHNOLOGISTS

Expanding upper midwest food processor has opportunities for two young Food Technologists with B.S. degrees in Food Technology or the equivalent in experience. Work involves quality control, development of new products and improvement of present products. Experience in canned and frozen food processing preferred.

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Opportunity also available for an experienced production supervisor in the canned and frozen food field. Duties include job assignments, quality control, safety and cost control.

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FOOD TECHNOLOGISTS

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Gladys Hunting (Consultant)
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The Heyworth Building
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29 E. Madison St., Chicago 2, Ill.

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Expanding Research Division of international food company has unusual opportunity for man with M.S. in Food Technology or B.S. plus some production or research experience in candy industry. Outstanding employee benefits. Salary open. REPLY BOX 765, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill.

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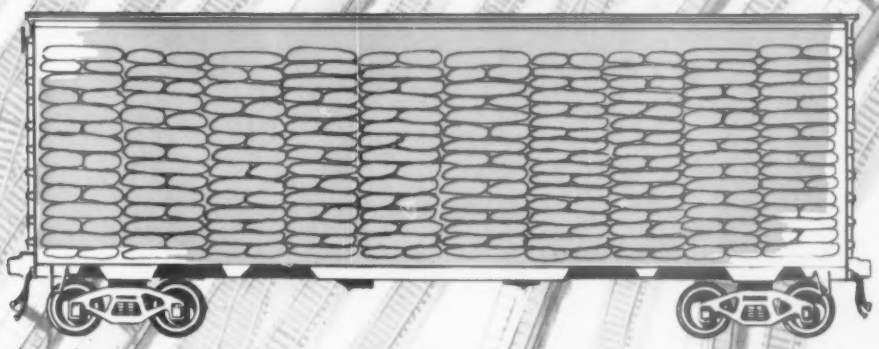

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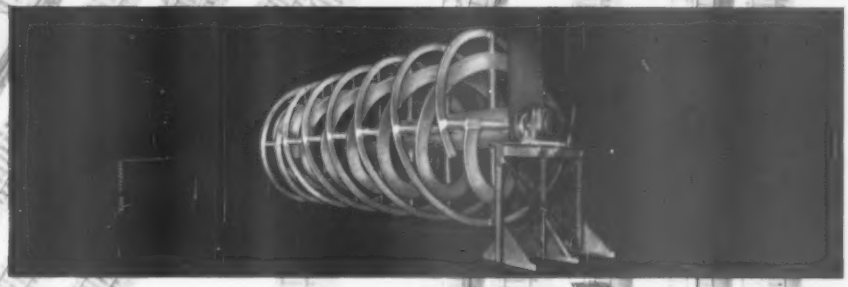

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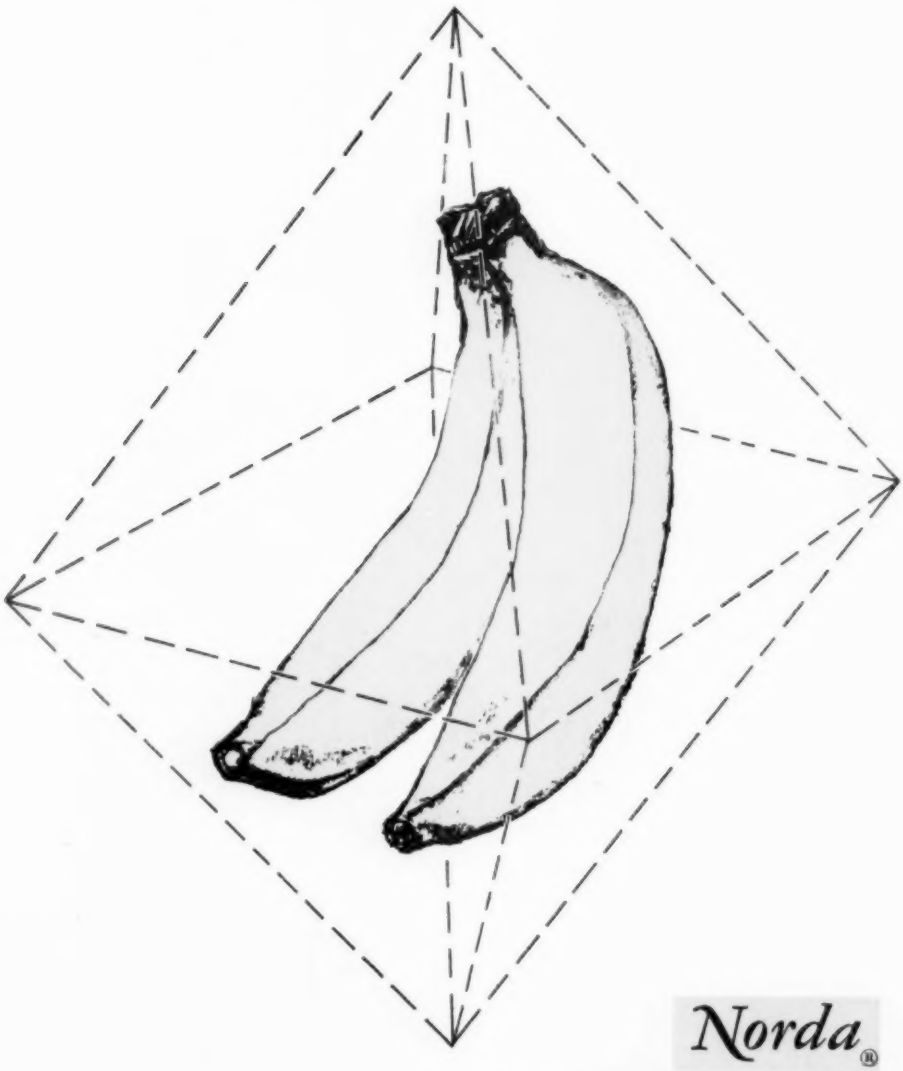

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